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A REVISION OF THE GENUS OMOPHRON (CARABIDAE, COLEOPTERA) OF NORTH AMERICA NORTH OF MEXICO[1]

CLARENCE A. BENSCHOTER AND EDWIN F. COOK

INTRODUCTION

Kirby (1837) is apparently the first worker to recognize this group as a family.[2] Leng (1920) and Böving and Craighead (1930), also recognize the family. The diagnostic characters used to separate the Omophronidae from the Carabidae are: form convex (almost hemispherical), oval and with the scutellum entirely concealed. Most Coleopterists consider this group a single genus, *Omophron*, of the tribe Omophronini of the Carabidae. With this latter opinion the authors concur.

Fabricius (1790) originally assigned the genus name *Scolytus* to this group, but since Geoffroy (1762) had already chosen this name for a group of bark beetles, Latreille (1802) created the new name *Omophron* and redescribed the genus.

Horn (1870) made the first attempt to revise the *Omophron* of North America and recognized 9 species. All of these remain valid. Casey (1897) felt that "a new tabular arrangement would appear to be desirable", and added 7 new species to the list. Casey described 14 species and 4 supspecies of *Omophron*, only 2 of which are considered by the author to be valid.

Bänninger (1921) considered all species for the world to be contained in the single genus *Omophron*, while Semenov-Tian-Shansky (1922) recognized 10 genera for the world, 4 of which include most of the species in North America north of Mexico.

This paper considers the species involved as belonging to the single genus *Omophron*.

The most serious omission in all of the foregoing works is the complete absence of illustration. In addition, no complete study of genitalia has ever been undertaken. In the world revisions (Bänninger, 1921 and Semenov-Tian-Shansky, 1922) the genitalia are mentioned briefly in an occasional species description, but not consistently, so that comparisons cannot be drawn throughout the group. The present study was undertaken in an attempt to alleviate this rather confusing situation by incorporating past and present findings into one work. Most of the original descriptions involve only external morphology. Hence many of the characters used were found to be common to all members of the genus and therefore of little diagnostic value at the specific level. Male genitalia proved to be quite useful and, in conjunction with other morphological characters, serve as the basis for the conclusions reached in this paper. The authors have observed little or no variation within a species. The shape of the apex of the aedeagus is one of the best characters for separating species, but will not distinguish between them all. In such situations, other morphological structures must be relied upon. In order that the group as a whole may be more clearly understood, it seems advisable to redescribe the genus as well as its component species.

Of the 29 species and 5 subspecies heretofore described for North America north of Mexico, the authors recognize 11 species. Subspecies are not recognized. Morphological variants are not geographically well defined (localized) and not sufficiently different taxonomically (Mayr et al., 1953). These beetles vary mainly in coloration of the elytra (ratio of dark to pale areas), and in the degree of metallic luster of the maculation; there is otherwise little variation within populations of the same species. The only cases in which clines seem to exist are in *O. americanus* and *O. robustus*.

[1]Paper No. 3395, Scientific Journal Series, Minnesota Agricultural Experiment Station, St. Paul.

Accepted for publication Sept. 23, 1955.

[2]Bonelli (1810) is cited by Jeannel (1941) as the first to use the family name. Examination of Bonelli's papers on Carabidae does not bear this out. *Omophron* Latreille is mentioned only as a synonym of *Scolytus* Fabricius.

Generally speaking, specimens of *O. americanus* show a reduction in maculation moving from east to west. For *O. robustus* the metallic green luster of the maculation decreases, also from east to west. In neither case is there any indication of a sharp break that would permit subdivision into subspecies.

Approximately 4500 specimens were examined during the course of this study. All type specimens deposited in The Academy of Natural Sciences of Philadelphia (2), and the U. S. National Museum (Smithsonian Institution) (18), were examined.

Very little is known of the biology of the genus. Both adults and larvae live in wet sand on the margins of lakes, rivers and streams. They are nocturnal, predacious, and apparently gregarious. The authors have never collected single isolated specimens. The ability to fly is controversial. Structurally, they are capable of flight but the authors have never seen one fly, nor been able to induce flight. A few specimens have been taken at light traps, so they probably can fly, but rarely do so. In Minnesota these beetles apparently over-winter as adults. Mating occurs in early spring and larvae can be collected throughout the summer. Both adults and larvae can be easily collected by pouring water over a sand bank. Dr. H. Dietrich (*in litt.*) and Mr. O. L. Cartwright (*teste*) state that large numbers can be taken at night with a flashlight. Repeated attempts were made to rear larvae in the laboratory but without success. In one instance a mature larva collected September 19 pupated September 23. The last larval skin was preserved. The adult beetle (*O. tesselatus*) emerged October 1. At this time the authors are unable to distinguish between species of *Omophron* larvae. Although they are not considered to be an economic pest, the larvae (according to Balduf, 1935) occasionally attack crop seedlings.

The following terminologies are used in this paper: Morphology, Blatchley (1910) and Torre-Bueno (1937); ♂ genitalia, Michener (1944) and Wood (1952); ♀ genitalia, Tanner (1927).

Omophron as a genus name was apparently considered to be neuter by the original describer. Csiki (1927) considered *Omophron* to be masculine and changed the specific endings accordingly. The authors were informed by Dr. Wm. A. McDonald of the Classical Languages Department, University of Minnesota, that *Omophron* is masculine in gender and, therefore, the endings as given by Csiki are correct.

METHODS

The genitalia of these beetles are entirely retracted into the abdominal cavity. It is virtually impossible to relax dried specimens sufficiently to completely extract the genitalia without damaging the specimen and/or the copulatory apparatus. Because of this, the entire specimen is first boiled for several minutes in distilled water to relax it; then the abdomen is removed and boiled in 10% KOH to remove all non-sclerotized material. The genitalia are then removed for closer scrutiny. The abdomen is placed on a point and the genitalia in a microvial of glycerin on the same pin with the specimen.

The following techniques were used in preparation of illustrative material. Structures which were small enough (genitalia, mouth parts, etc.) were mounted on slides in Canada balsam and drawn with the aid of a microprojector. The larger structures (elytra, heads, pronota, abdomen, etc.) were imbedded in glycerin jelly, a temporary mount, and drawn with the aid of an Eberbach scale projection machine.

For loan of material and other assistance, the authors wish to thank Dr. G. E. Ball, University of Alberta, Dr. W. F. Barr, University of Idaho, Dr. R. H Beamer, University of Kansas, Dr. E. C. Becker, Canadian National Collections, Dr. M. A. Cazier, American Museum of Natural History, Dr. F. G. Clarke and Mr. O. L. Cartwright, U. S. National Museum (Smithsonian Institution), Prof. J. J. Davis, Purdue University, Dr. H. Dietrich, Cornell University, Mr. W. R. Enns, University of Missouri, Dr. M. H. Hatch, University of Washington, Dr. L. A. Hetrick, University of Florida, Dr. P. D. Hurd, Jr., University of California (Berkeley), Dr. M. T. James, Washington State College, Dr. H. King, Michigan State College, Dr. J. L. Lafoon, Iowa State College, Dr. F. A. Lawson, Kansas State College, Mr. H. B. Leech, California Academy of Sciences, Mr. B. Malkin, University of Washington, Mr. A. T. McClay, University of California (Davis), Dr. L. W. Quate, University of Nebraska, Mr. J. A. G. Rehn, The Academy of Natural Sciences of Philadelphia, Dr. H. J. Reinhard, Texas A & M College, Mr. V. D. Roth, Oregon State College, Dr. M. W. Sanderson, Illinois State Natural History Survey, Dr. R. D. Shenefelt, University of Wisconsin, Dr. M. E. Smith, University of Massachusetts, and Dr. V. M. Tanner, Brigham Young University.

Genus **Omophron** Latreille[3]

Scolytus Fabricius, 1790. Naturh.-selsk., Copenhagen, Skrivter, 1: 221 (preoccupied by *Scolytus* Goeffroy, 1762).

Omophron Latreille, 1802. Hist. Nat. Crust. Ins., 3: 89.

Homophron Rye, 1875. Zool. Rec., 12: 279. (Emendation).

Homophron Semenov-Tian-Shansky, 1922. Rev. Russe. d'Ent., 18: 41 (Type: *O. tesselatus* Say).

Istor Semenov-Tian-Shansky, 1922. Rev. Russe. d'Ent., 18: 43. (Type: *O. robustus* Horn).

Paromophron Semenov-Tian-Shansky, 1922. Rev. Russe. d'Ent., 18: 40. (Type: *O. ovalis* Horn).

Prosencon Semenov-Tian-Shansky, 1922. Rev. Russe. d'Ent., 18:44. (Type: *O. gilae* Leconte).

[3]For a complete bibliography of this group through 1927, consult E. Csiki, 1927, Carabidae I, Col. Cat.—W. Junk, 1: 404–412.

Type Species: **Carabus limbatus** Fabricius, 1776. (European)

Semenov-Tian-Shansky divided this group into 10 genera for the world. Most, but not all, of the North American species were included in his 4 genera above. *O. labiatus, obliteratus* and *grossus* apparently were not included, at least they are not mentioned in the generic descriptions. Investigation of Semenov's paper reveals that some of his genera were separated by only a single character. If the interpretation is correct he divides the North American genera as follows:

Elytra 15-striate = *Paromophron* and *Homophron*
Elytra 14- striate = *Istor* and *Prosencon*

These are further subdivided as follows.

Paromophron (Type: *O. ovalis*) = middle coxae bearing a single seta.

Homophron (Type: *O. tesselatus*) = middle coxae with accessory setae in addition to regular seta.

The former is apparently a monotypic genus while the latter includes *tesselatus, americanus, dentatus, solidus* and *nitidus*. According to the findings of the authors, *ovalis* is morphologically more closely related to the other species of *Homophron* than is *nitidus*. In other words if any of these species were to be put into a separate genus it should be *nitidus*.

Istor (Type: *O. robustus*) = epimeron and episternum of prosternum divided.

Prosencon (Type: *O. gilae*) = epimeron and episternum of prosternum not divided.

Examination of the prothorax of specimens of the type species of these two so-called genera reveals no differences. There is a pleural suture between proepimeron and proepisternum in both species. Thus, these genera are separated by Semenov on the basis of 2 valid but minor characters. The characters used for separating 2 of these genera are based on an observational error. The valid characters seem to the authors to be specific rather than generic. The North American species are exceedingly homogeneous in all other characters. We have, therefore, chosen to place the 4 names proposed for the north American species in synonymy.

DESCRIPTION

The characters which distinguish this genus from all other Carabidae are: Scutellum apparently absent (at least entirely concealed) and convex (almost hemispherical), oval shape. The species range in mean length from 4.4–9.0 mm.

Head: Head oval, fitting closely into prothorax. Dorsal surface punctate. Front forming a prominent ridge over each antennal pit and bearing a seta on each lateral margin mesal to eyes. Clypeus with a seta near each lateral margin (Fig. 2-C). Genal area on ventral surface of head impressed and usually punctate. Gula with either 2 or 4 setae (Fig. 2-K, L). Antenna filiform, 11-segmented with proximal 4 segments glabrous (except for apical setae), distal 7 segments pubescent. Basal segment much larger than others and bearing a single prominent seta (Fig. 1-H). Labrum rectangular in shape with 6 large setae on dorsal anterior margin. Mandible curved and acute at tip. Two or three simple teeth are present on mesal margin near the base. A median ridge divides dorsal surface, the lateral portion of which bears a single large seta. Maxillary palpus 4-segmented, apex of distal segment truncate; galea slender and 2-segmented; lacinia slender with a curved acute apex and bearing strong bristles on its inner margin. Labial palpus 3-segmented, basal segment short and glabrous, middle segment long and bearing a row of long setae, distal segment glabrous with apex truncate. Ligula with 2 setae near midline. Mouth parts are figured in Fig. 1.

Thorax: Anterior margin of pronotum broadly emarginate to accommodate the head, posterior margin sinuate (Figs. 2-E, J). Marking on dorsal surface of pronotum is a very important taxonomic character for this group. Prosternum with a file-like surface on mesal anterior margin. This may be a stridulating organ. Prosternum punctate and prolonged posteriorly into a shovel-shaped process. Thoracic pleura usually punctate. General color of pleura and sternites testaceous to ferrugineous. All tarsi 5-segmented, each with 2 claws on last segment. Basal segment of protarsus expanded in ♂ and bearing a ventral pad of short setae. Second tarsal segment may also bear a ventral pad of setae (Fig. 1-E). Basal segment of mesotarsus not expanded in the ♂ but also bearing a ventral pad of setae (Fig. 1-F). Tibiae of prothoracic legs with inner surface emarginate at distal ends. No setae apparent on procoxae; 2 setae on each trochanter of prothoracic legs; 1 seta on each trochanter of mesothoracic legs. Elytra covering abdomen entirely. Dorsal maculation consisting in general of 3 dark irregular fasciae or transverse bands on a lighter ground color. These fasciae may be continuous, but are most commonly broken. The sculpture of each elytron consists of 14 or 15 longitudinal furrows or striae containing punctures. Epipleuron with a tooth-like process on inner margin.

Abdomen: With 6 visible sternites, a pair of ambulatorial setae on fourth and sixth sternites (Fig. 4-F). General color of ventral surface testaceous to ferrugineous. Eighth sternite (retracted) divided in ♀. Female genitalia are similar for all species. They are very simple, consisting only of a pair of sclerites, here termed the coxites, each of which bears a short stylus (Fig. 4-D). Male genitalia are basically similar in structure, consisting of gonocoxites, 2 asymmetrical gonostyli, and the aedeagus (penis and fused penis valves) (Fig. 5-B). According to Wood (1952), the gonocoxites are fused. However, in at least one species, *americanus*, they

appear to be distinct. The shape of the apex of the aedeagus is one of the most important taxonomic characters for this group.

KEY TO THE SPECIES OF OMOPHRON OF NORTH AMERICA NORTH OF MEXICO

1. Elytron 14-striate, all striae distinct 2
   Elytron 15-striate, striae may be indistinct near lateral margin 4
2. (1) Pale area on front V-shaped (as in Fig. 2-B), pronotum generally dark with pale lateral margins (as in Plate 2-G) . . . . . **obliteratus** Horn
   Pale area on front M-shaped (as in Fig. 2-D), pronotum generally pale with a discal spot or narrow transverse band (Fig. 2-F) 3
3. (2) Elytral striae deep and closely punctured, 2 setae on metacoxa (as in Fig. 4-B), luster dull . . . . . **gilae** Lec.
   Elytral striae feebly impressed with punctures large and widely separated, 1 seta on metacoxa (Fig. 4-C), luster shining . . . **robustus** Horn
4. (1) Pronotum entirely dark with the pale area on the lateral margins wide and even in outline (as in Fig. 2-E), form broadly oval, the sides of the elytra and pronotum forming a continuous arc 5
   Pronotum, if predominantly dark, always with pale area on lateral margins irregular in outline (as in Fig. 2-G), form less oval, the sides of the elytra and pronotum not continuous in curvature 6
5. (4) General color of dorsal surface a non-metallic reddish-brown to brownish-black, elytra entirely dark except for a narrow pale marginal area expanded toward the apex (Fig. 3-G), punctures indistinct toward lateral margins . . . . . **labiatus** (Fabr.)
   General color above dark with a metallic green luster, dark areas of elytra show evidence of forming 3 transverse bands (Fig. 3-H), striae faint, punctures large and distinct at lateral margins . . . . . **nitidus** Lec.
6. (4) Front with pale area variable but not distinctly V-shaped 7
   Front with pale area distinctly V-shaped (as in Fig. 2-B) 9
7. (6) Pronotum with an irregular transverse band (as in Fig. 2-I), pale area on front weakly M-shaped (Fig. 2-C) . . . . . **dentatus** Lec.
   Pronotum with a spot on median third (as in Fig. 2-J) 8
8. (7) Total length[4] less than 7.5 mm., pale area on front distinctly M-shaped (Fig. 2-D) . . . . . **tesselatus** Say
   Total length greater than 7.5 mm., frontal area entirely pale (Fig. 2-A) . . . . . **grossus** Csy.
9. (6) Dark spot of pronotum forming an irregular transverse band which joins the posterior margin in 2 or 3 places (Fig. 2-I) . . **solidus** Csy.
   Dark spot of pronotum broad on posterior margin (as in Fig. 2-G) 10
10. (9) Metasternum with disc impunctate, 1 seta on mesocoxa (as in Fig. 4-A), 2 gular setae (Fig. 2-L), 5th abdominal sternite most commonly bearing a pair of ambulatorial setae (Fig. 4-G) . . . . . **ovalis** Horn
    Metasternum with disc punctate, 2 setae on mesocoxa (as in Fig. 4-E), 4 gular setae (as in Fig. 4-E), 5th abdominal sternite usually without paired ambulatorial setae (as in Fig. 4-F) . . . . . **americanus** Dej.

[4]Total length equal to distance from base of the antenna to apex of the elytra, with the beetle measured in lateral view.

Omophron obliteratus Horn

Omophron obliteratum Horn, 1870. Trans. Amer. Ent. Soc., 3: 73.
Omophron sonorae Casey, 1897. Ann. New York Acad. Sci., 9: 304.
Omophron obliteratum utense Casey, 1913. Mem. Col., IV, p. 43.
Omophron obliteratum subimpressum Casey, 1913. Mem. Col. IV, p. 43.
Omophron obliteratus Horn. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 405.

Holotype: Sex ♀, repository, The Academy of Natural Sciences of Philadelphia, Type No. 1011 examined. *Type locality:* Camp Grant on the San Pedro River, a tributary of the Gila, Arizona.

DESCRIPTION

MALES: Total length 5.9–6.9 mm. (M 6.4 mm.), width 3.8–4.3 mm. (M 4.1 mm.), l/w ratio 1.48–1.63 (M 1.55).

FEMALES: Total length 5.9–7.3 mm. (M 6.7 mm.), width 3.9–4.9 mm. (M 4.4 mm.), l/w ratio 1.45–1.60 (M 1.54).

Head: Pale area on front V-shaped (as in Fig. 2-B); dorsal surface punctate; usually with only 2 gular setae (as in Fig. 2-L).

Thorax: Pronotum predominantly dark with pale area on lateral margins irregular in outline (as in Fig. 2-G); dorsal surface coarsely and sparsely punctured; disc of metasternum punctate.

Thoracic appendages: Elytron 14-striate, with punctures large and widely spaced, becoming obliterated toward apical third; maculation dark and extensive, exceeding pale ground color in area. Mesocoxa and metacoxa each with 2 setae (as in Figs. 4-B, E).

Abdomen: A pair of ambulatorial setae (often difficult to see) on 5th sternite (as in Fig. 4-G). Penis of ♂ with a very small knob-like structure approximately at mid point of ejaculatory duct; aedeagus elongate with sides parallel, apex broadly truncate (Fig. 5-J).

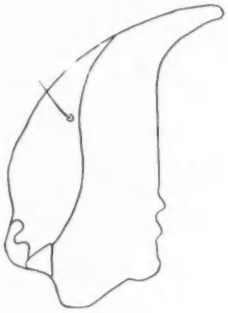
COMMENTS: This species superficially looks very much like *americanus* in its dark coloration. However, it is slightly larger and more elongate than the latter. The diagnostic characters which best separate it from all other species are: 14-striate elytron, V-shaped light area on front, and structure of the ♂ genitalia.

O. obliteratus occurs in the southwest United States, its known distribution including Texas,

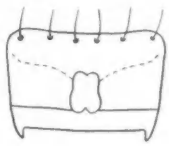
EXPLANATION OF FIGURE I

O. tesselatus

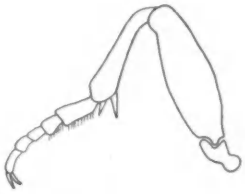
A. Left mandible, dorsal aspect. B. Left maxilla, dorsal aspect. C. Labium, ventral aspect. D. Labrum, dorsal aspect. E. Prothoracic leg of ♂. F. Mesothoracic leg of ♂. G. Metathoracic leg of ♂. H. Right antenna, dorsal aspect. (Pubescence on distal 7 segments omitted.)



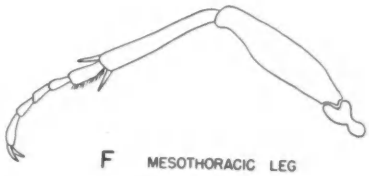
A MANDIBLE



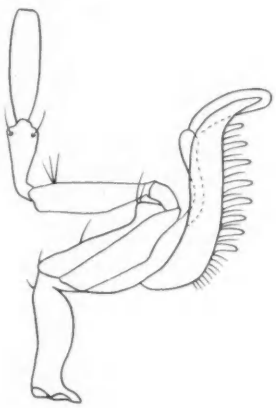
D LABRUM



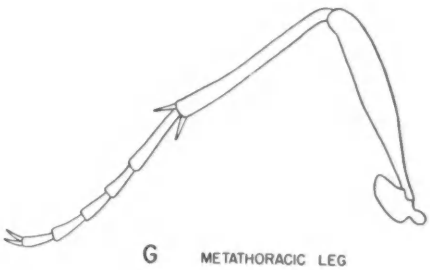
E PROTHORACIC LEG



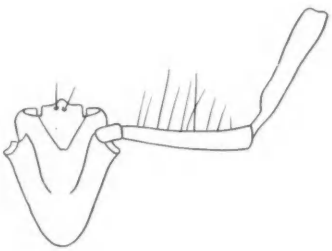
F MESOTHORACIC LEG



B MAXILLA



G METATHORACIC LEG



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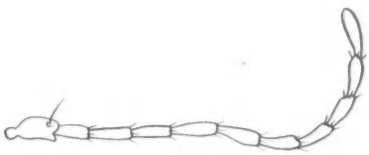


H ANTENNA

O. TESSELATUS

New Mexico, Utah, Arizona, southern California and Mexico.

SPECIMENS EXAMINED: *Holotype; O. sonorae* Casey, ♀; Sonora, Mexico. Repository, U. S. National Museum, Type No. 48091. Also 3 ♂♂ paratypes labelled Sonora, deposited with holotype. *Holotype: O. obliteratum utense* Casey, ♀; Leeds, Utah; August 4–7. Repository, U. S. National Museum, Type No. 48095. Also 2 ♀♀ paratypes labelled St. George, Utah; July. Deposited with holotype. *Holotype: O. obliteratum subimpressum* Casey, ♀; New Mexico. Repository, U. S. National Museum, Type No. 48096.

TEXAS: Limpia Canyon (Davis Mts.); July 14–22; NEW MEXICO: Grant Co.; July 3 to August 4; UTAH: Chad's Rch., Kolob, St. George, Zion Nat'l Park; July; ARIZONA: S. Arizona, Bill Williams Fork, Dragoon (Cochise Co.), Cottonwood, Douglas, Globe, Nogales, Oak Creek Canyon (near Flagstaff), Patagonia, Phoenix, Pima Co., Prescott, Roosevelt Lake, Santa Cruz Co. (5 mi. S. W. Patagonia), Tucson; April 4 to November 7; CALIFORNIA: So. California; no date; MEXICO—CHIHUAHUA: Santa Barbara (63 miles west), Huejotitlan; July 20–21; DURANGO: San Juan del Rio; July 30; SONORA: Hermosillo; April 19; TAMPAULIPAS: Matamoros; May.

Total Specimens Examined: 398.

Omophron gilae Leconte

Omophron gilae Leconte, 1852. Ann. Lyceum Nat. Hist. New York, 5: 201.
Omophron pallidum Casey, 1897. Ann. New York Acad. Sci., 9: 305.
Omophron gilae pimale Casey, 1913. Mem. Col., IV, p. 44.

HOLOTYPE: Sex (?), repository, Museum of Comparative Zoology, Harvard College. *Type locality:* Gila River, Arizona.

DESCRIPTION

MALES: Total length 4.4–6.4 mm. (M 5.8 mm.), width 3.3–4.3 mm. (M 4.0 mm.), l/w ratio 1.39–1.52 (M 1.45).

FEMALES: Total length 5.7–6.9 mm. (M 6.2 mm.), width 3.9–4.7 mm. (M 4.3 mm.), l/w ratio 1.41–1.53 (M 1.45).

Head: Pale area on front M-shaped (as in Fig. 2-D); dorsal surface coarsely and sparsely punctured; with only 2 gular setae (as in Fig. 2-L).

Thorax: Pronotum with a spot on median ⅓ (Fig. 2-H); dorsal surface coarsely and sparsely punctured; disc of metasternum punctate.

Thoracic appendages: Elytron 14-striate, striae deep with convex intervals, punctures large and widely spaced; maculation rather sparse, exceeded in area by pale ground color. Mesocoxae and metacoxae each with 2 setae (as in Fig. 4-B, E).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (as in Fig. 4-F). Male genitalia with ejaculatory duct of penis simple; apex of aedeagus acute (Fig. 5-E).

COMMENTS: This light-colored beetle at first glance might be confused with *robustus* and *tesselatus*. The diagnostic characters which will readily distinguish it are: 14 striate elytra, 2 setae on the metacoxa, and the structure of the ♂ genitalia.

O. gilae occurs in the southwest United States, its known distribution including Texas, New Mexico, Colorado, Utah, Arizona and southern California.

SPECIMENS EXAMINED: *Holotype: O. pallidum* Casey, ♂; southwestern Utah. Repository, U. S. National Museum, Type No. 48093. Also 1 ♀ paratype labelled Utah. Deposited with holotype. *Holotype: O. gilae pimale* Casey, ♂; Arizona. Repository, U. S. National Museum, Type No. 48092.

TEXAS: No locality or date; NEW MEXICO: No locality or date; COLORADO: Glenwood Springs; August; UTAH: St. George, Santa Clara, Washington Co.; May 9 to July 28; ARIZONA: Dome, Ehrenberg, Gila Bend, Gila River, Laguna, Palomas, Phoenix, Pima Co. (Quitobaquito Organ Pipe Nat'l Mon.), Riverside, Salt River, Tempe, Yuma; June 14 to August 20; CALIFORNIA: Blythe (Riverside Co.), Colorado River (Blythe); April 12 to August 15.

Total Specimens Examined: 132.

Omophron robustus Horn

Omophron robustum Horn, 1870. Trans. Amer. Ent. Soc., 3: 74.
Omophron brevipenne Casey, 1909. Canad. Ent., 41: 276.
Omophron decoloratum Fall, 1920. Canad. Ent., 52: 211. NEW SYNONOMY.
Omophron decoloratus Fall. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 406.
Omophron robustus Horn. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 411.

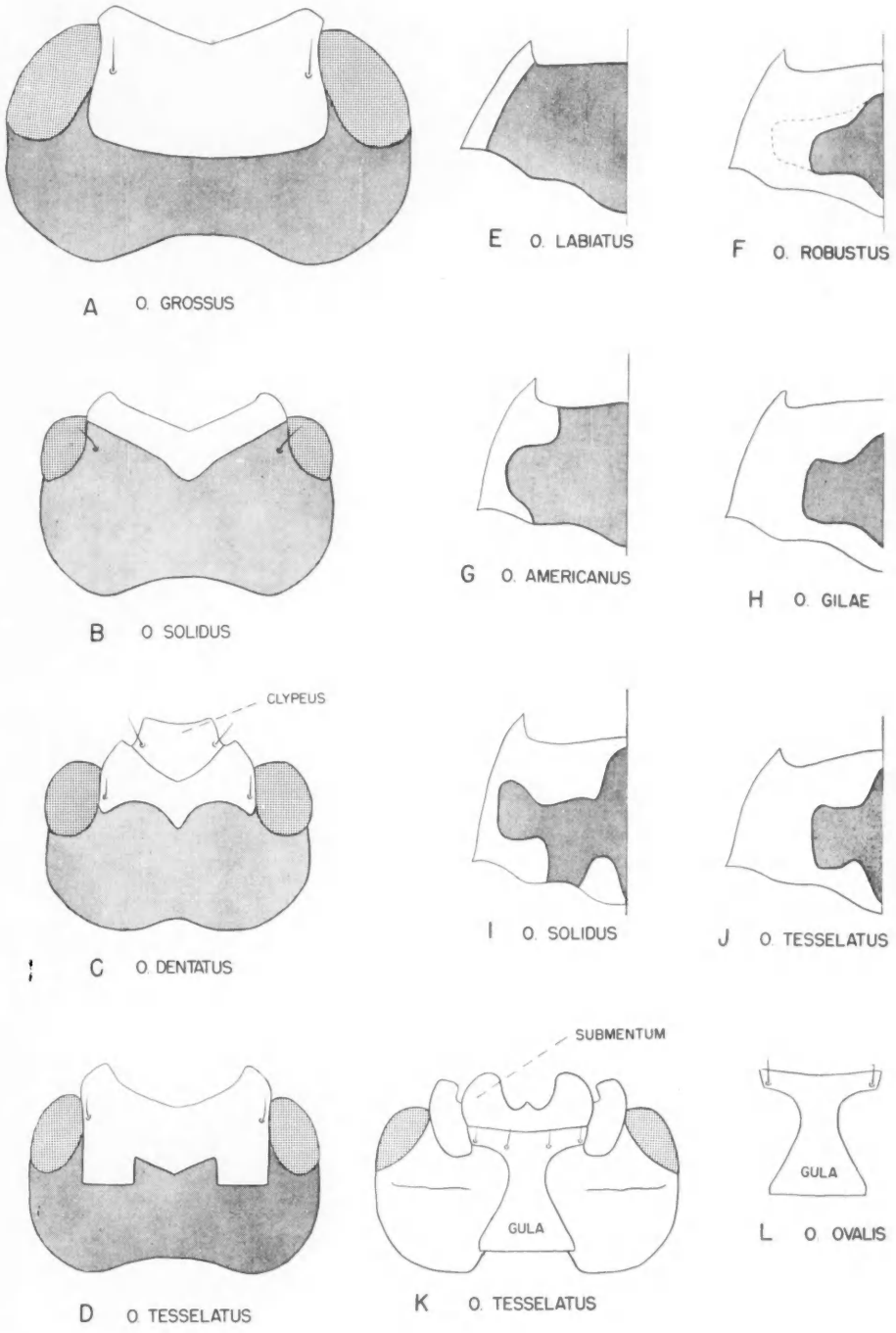
HOLOTYPE: Sex (?), repository, Museum of Comparative Zoology, Harvard College. *Type locality:* Nova Scotia region.

DESCRIPTION

MALES: Total length 5.2–6.4 mm. (M 6.0 mm.), width 3.8–4.7 mm. (M 4.3 mm.), l/w ratio 1.35–1.42 (M 1.39).

EXPLANATION OF FIGURE II

A. Head of *O. grossus*, dorsal aspect. B. Head of *solidus*, dorsal aspect. C. Head of *dentatus*, including clypeus, dorsal aspect. D. Head of *tesselatus*, dorsal aspect. E. Pronotum of *labiatus*, dorsal aspect. F. Pronotum of *robustus*, dorsal aspect. (Dotted line indicates maximum extent of maculation.) G. Pronotum of *americanus*, dorsal aspect. H. Pronotum of *gilae*, dorsal aspect. I. Pronotum of *solidus*, dorsal aspect. J. Pronotum of *tesselatus*, dorsal aspect. K. Head of *tesselatus*, ventral aspect, showing position of 4 gular setae. L. Gula of *ovalis* showing position of 2 gular setae.


A O. GROSSUS
E O. LABIATUS
F O. ROBUSTUS
B O. SOLIDUS
G O. AMERICANUS
H O. GILAE
CLYPEUS
C O. DENTATUS
I O. SOLIDUS
J O. TESSELATUS
SUBMENTUM
GULA
D O. TESSELATUS
GULA
K O. TESSELATUS
L O. OVALIS

FEMALES: Total length 5.3–6.9 mm. (M 6.3 mm.), width 3.9–5.1 mm. (M 4.5 mm.), 1/w ratio 1.34–1.51 (M 1.40).

Head: Pale area on front M-shaped (as in Fig. 2-D); dorsal surface coarsely and sparsely punctured; with 2 gular setae (as in Fig. 2-L).

Thorax: Pronotal marking varies from a discal spot to a transverse band (Fig. 2-F); punctures on dorsal surface generally confined to anterior and posterior margins, with few if any on discal area; disc of metasternum punctate.

Thoracic appendages: Elytron 14-striate, striae feebly impressed with large widely-spaced punctures; maculation sparse, most often exceeded in area by the pale ground color. Mesocoxae and metacoxae each with only 1 seta (Fig. 4-A, C).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (as in Fig. 4-F). Penis of ♂ with a knob-like projection approximately at midpoint of the ejaculatory duct; apex of aedeagus acute (Fig. 5-F).

COMMENTS: This species shows great variation in the color of the maculation. How much of this is due to fac ing of the specimens is difficult to determine, but an east west color cline seems to exist. Specimens of the eastern extreme of the known range (Ohio and Ontario) tend to have the maculation bright metallic green in color while those on the western extreme (Alberta) have the maculation somewhat less extensive and almost completely without metallic green color. This is not a clear cut situation of course, but the trend is there. Diagnostic characters are: broadly oval form, 14-striae, only 1 seta on metacoxa, and structure of the ♂ genitalia. *O. robustus* is a widely distributed species, its known range including Ontario, Michigan, Ohio, Indiana, Illinois, Wisconsin, Minnesota, Iowa, Oklahoma, Kansas, Nebraska, North Dakota and Alberta.

SPECIMENS EXAMINED: *Holotype: O. brevipenne* Casey, ♀; Ohio. Repository, U. S. National Museum, Type No. 48087.

ONTARIO: Prince Edward Co., Toronto, Walkerville; May 7 to August 14; MICHIGAN: Detroit, July 4, Monroe (no date); OHIO: No locality or date; Cincinnati; June 8; INDIANA: No locality or date; ILLINOIS: No locality or date; WISCONSIN: Dane Co.; July 6; MINNESOTA: Crow Wing Co., Garrison, Mille Lacs Lake, Nicollet Co.; May 14 to July 13; IOWA: Ames, Boone, Solon; May 1 to August 21; OKLAHOMA: Alfalfa Co., Great Salt Plains (Alfalfa Co.), Greer Co.; June 6 to July 24; KANSAS: Blackjack Creek (Pottawatomie Co.), Graham Co., Gray Co., Pottawatomie Co., Wallace Co.; June 7 to August 16; NEBRASKA: Ashland, Clear Lake (Cherry Co.), Lincoln, West Point; April 26 to July 16; NORTH DAKOTA: Devil's Lake, Mission Lake; June 29 to July 4; ALBERTA: Chappice Lake, Medicine Hat; July 29 to August 29.

Total Specimens Examined: 189.

Omophron labiatus (Fabricius)

Scolytus labiatus Fabricius, 1801. Syst. Eleuth., 1: 248. *Omophron labiatus* (Fabricius). Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 407–408.

HOLOTYPE: Sex (?), repository unknown to author. *Type locality:* Carolina.

DESCRIPTION

MALES: Total length 4.9–5.8 mm. (M 5.4 mm.), width 3.6–4.3 mm. (M 3.9 mm.), 1/w ratio 1.34–1.44 (M 1.37).

FEMALES: Total length 4.8–6.3 mm. (M 5.8 mm.), width 3.4–4.6 mm. (M 4.2 mm.), 1/w ratio 1.35–1.45 (M 1.39).

Head: Pale area on front V-shaped (as in Fig. 2-B); dorsal surface finely but sparsely punctured; with 4 gular setae (as in Fig. 2-K).

Thorax: Pronotum broad at base forming a continuous arc with margins of elytra, dorsal surface entirely dark with pale areas on lateral margins wide and even in outline (Fig. 2-E). Punctures fine and sparse, and more or less limited to the periphery of the pronotum; disc of metasternum punctate.

Thoracic appendages: Elytron apparently 15-striate, the striae and punctures both becoming indistinct or obliterated toward apex and lateral margins. Elytra entirely dark, except for a narrow light marginal area which becomes expanded toward apices. Maculation shows no evidence of forming 3 transverse bands (Fig. 3-G). Mesocoxae and metacoxae each with 2 setae (as in Figs. 4-B, E).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (as in Fig. 4-F). Male genitalia with lateral margins of aedeagus slightly convergent near apex; apex narrowly truncate (Fig. 5-G).

COMMENTS: *O. labiatus* is distinctive enough so that once seen, it can never be confused with any of the other species of this group. Structurally, it is most closely related to *nitidus*, but averages larger in size. The general color of the dorsal surface ranges from reddish-brown to brownish-black. The diagnostic characters for this species are: broadly oval form, pale area on lateral margins of pronotum even in outline, elytral striae and punctures becoming indistinct or obliterated toward lateral margins, maculation

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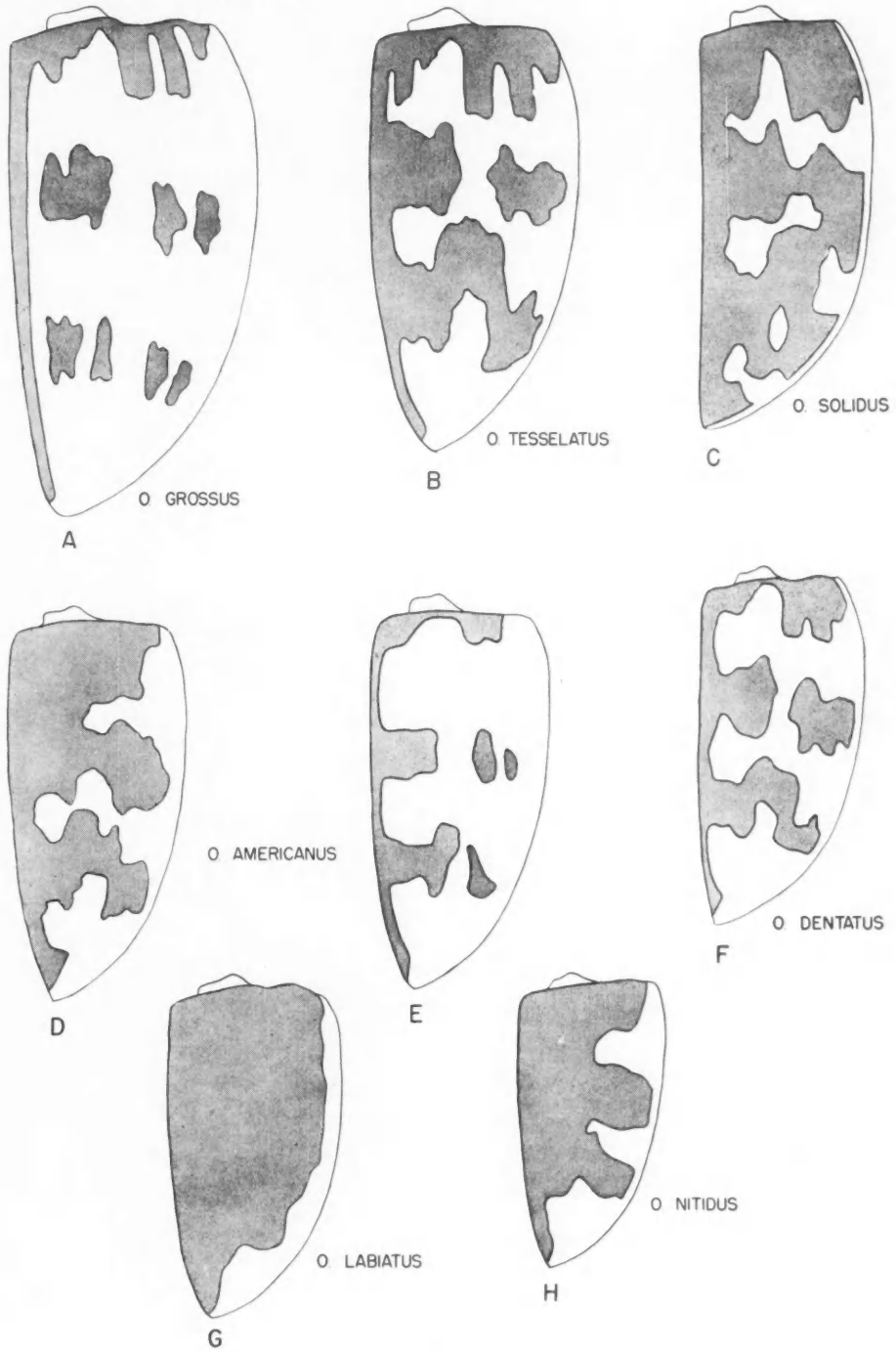
EXPLANATION OF FIGURE III

Typical Elytral Maculation
(Striae and Punctures Omitted)

A. Right elytron, *O. grossus*. B. Right elytron, *tesselatus*. C. Right elytron, *solidus*. D. Right elytron, *americanus*, showing typical pattern of individuals in the northeastern United States. E. Right elytron, *americanus*, showing maximum reduction of maculation on western extreme of the range (Utah). F. Right elytron, *dentatus*. G. Right elytron, *labiatus*. H. Right elytron, *nitidus*.


O. GROSSUS
A
O. TESSELATUS
B
O. SOLIDUS
C
O. AMERICANUS
D
E
O. DENTATUS
F
O. LABIATUS
G
O. NITIDUS
H

of dorsal surface very extensive, not forming 3 transverse bands; dark color without metallic green luster, and structure of the ♂ genitalia.

This species shows a very interesting distribution, and appears to inhabit primarily the coastal states extending from New York to Florida to Texas. The known range includes New York, New Jersey, Pennsylvania, Maryland, District of Columbia, Virginia, Georgia, Florida, Alabama, Mississippi, Louisiana and Texas.

SPECIMENS EXAMINED: NEW YORK: Babylon, Jones Beach, Long Beach (L.I.); June 21 to September 30; NEW JERSEY: Anglesea, Atlantic City, Atsion, Brookville, Clementon, Collingswood, Cramer Hill, Hopatcong, Lakehurst, So. New Jersey, Westville, Wildwood, Woodbury; April 20 to July 29; PENNSYLVANIA: Philadelphia, So. Philadelphia; April 18 to June; MARYLAND: No locality or date; DISTRICT OF COLUMBIA: No locality or date; VIRGINIA: No locality or date; GEORGIA: Billy's Island (Okefenokee Swamp), Brunswick, St. Simon's Island, Spring Creek (Decatur Co.), Thomasville; May 3 to August 28; FLORIDA: Arcadia, Biscayne Bay, Brighton (Okeechobee), Brookville, Bunker Branch (Arcadia), Capron, Clearwater, Dunedin, Enterprise, Ft. Lauderdale, Ft. Myers, Gainesville, Hillsborough Co., Indian River, LaBelle, Lacoochee, Lake Okeechobee (So. Bay), Lake Placid, Lake Worth, Miami, Moore Haven, Odessa, Okeechobee, Onesco (Manatee Co.), Pablo Beach, Palatka, Punta Gorda, St. Petersburg, Sand Pt., Sanford, Sebring, Stuart, Tampa, Winter Park; January 20 to November; ALABAMA: Mobile; no date; MISSISSIPPI: Lucedale, Pass Christian; June 20–25; LOUISIANA: New Orleans; May 30; TEXAS: Corpus Christi, Galveston; April 29 to May 26.

Total Specimens Examined: 349.

Omophron nitidus Leconte

Omophron nitidum Leconte, 1848. Ann. Lyceum Nat. Hist., New York, 4: 447.
Omophron nitens Chaudoir, 1868. Rev. Mag. Zool., [2] 20: 60.
Omophron nitidus Leconte. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 410.

HOLOTYPE: Sex (?), repository, Museum of Comparative Zoology, Harvard College. *Type locality:* Missouri region.

DESCRIPTION

MALES: Total length 4.4–5.5 mm. (M 5.0 mm.), width 3.1–4.0 mm. (M 3.6 mm.), l/w ratio 1.34–1.45 (M 1.39).

FEMALES: Total length 4.6–5.7 mm. (M 5.3 mm.), width 3.3–4.2 mm. (M 3.9 mm.), l/w ratio 1.33–1.43 (M 1.38).

Head: Pale area on front V-shaped (as in Fig. 2-B); dorsal surface coarsely punctured; with 4 gular setae (as in Fig. 2-K).

Thorax: Pronotum broad at base, forming a continuous arc with margins of elytra, dorsal surface entirely dark with a bright green metallic luster. Pale areas on lateral margins wide and even in outline (as in Fig. 2-E). Punctures mainly peripheral; disc of metasternum punctate.

Thoracic appendages: Elytron 15-striate, striae feebly impressed; punctures very large, widely spaced and distinct to lateral margins but becoming obliterated toward apex; dark area predominating and showing evidence of forming 3 transverse bands (Fig. 3–H). Mesocoxae and metacoxae each with 2 setae (as in Figs. 4-B, E).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (as in Fig. 4-F). Male genitalia with aedeagus relatively short and broad, sides parallel, apex broadly truncate (Fig. 5-H).

COMMENTS: This beetle is the smallest member of the genus in North America north of Mexico. Structurally, it is closely related to *labiatus*. The general coloration of the dorsal surface is dark with a bright green or bluish-green metallic luster. *O. nitidus* is best distinguished from the other species by its broadly oval form, light area of lateral margins of pronotum even in outline, 15-striae with punctures very large, widely spaced and distinct at lateral margins, maculation with a bright metallic luster and showing evidence of forming 3 transverse bands, and structure of the ♂ genitalia.

This species is most common in the central states and has been recorded for Minnesota, Nebraska, Iowa, Illinois, Indiana, Tennessee, Missouri, Kansas, Oklahoma, Texas, Arkansas, Louisiana, Mississippi, and Alabama.

SPECIMENS EXAMINED: MINNESOTA: Hennepin Co., Ramsey Co.; no dates; NEBRASKA: Hogans Bridge (Rock Co.), Holt Co.; August 3 to September 1; IOWA: Ames; May 14; ILLINOIS: No. Illinois; no date; INDIANA: Lake Co., Pine; April 23 to May 5; TENNESSEE: Clarksville, Nashville; July 5; MISSOURI: Kansas City, St. Louis; July 30; KANSAS: Big Turkey Creek (Chanute), Douglas Co., Drury, Ellsworth Co., Ft. Scott, W. Kansas, Lawrence, Manhattan, McPherson, Medora (Sand Dunes), Neosho Co., Neosho River (Chanute), Ottawa, Rags, Saline Co., Sedwick Co., Topeka; April 7 to October 19; OKLAHOMA: Adair Co., Beckham Co., Hansen, Indian Territory, Major Co., Muskogee Co., Okmulgee, Pottawatomie Co., Tillman Co., Tulsa, Wichita Nat'l Forest, Wister; June 15 to August

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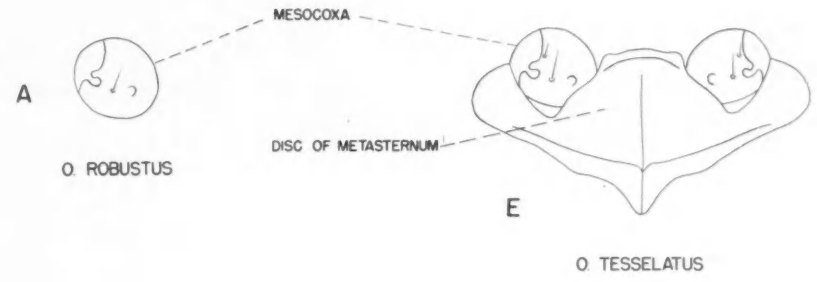
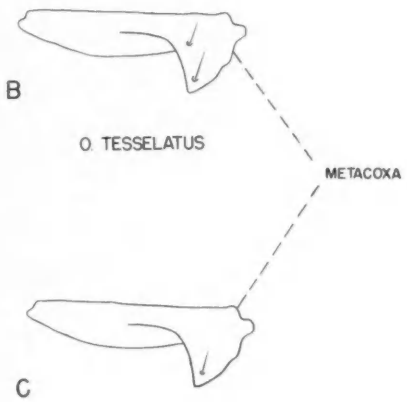
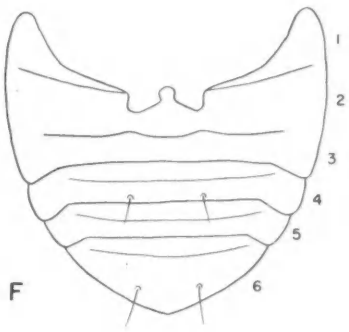
EXPLANATION OF FIGURE IV

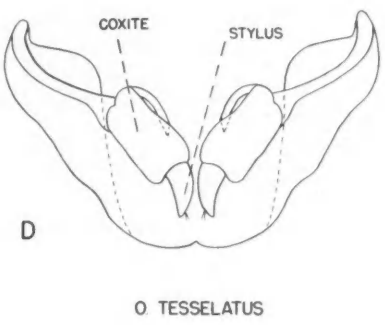
Ventral Sclerites

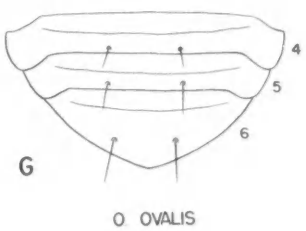
A. Mesocoxa of *robustus* bearing a single seta. B. Metacoxa of *tesselatus* bearing 2 setae. C. Metacoxa of *robustus* bearing a single seta. D. Female genitalia of *tesselatus*, ventral aspect. E. Mesocoxae bearing 2 setae, and discal region of metasternum, *tesselatus*. F. Abdomen of *tesselatus*, ventral aspect, showing 6 visible sternites with paired ambulatorial setae on 4th and 6th. G. Abdominal sternites 4–6, all bearing paired ambulatorial setae, *ovalis*.


MESOCOXA
A
DISC OF METASTERNUM
O. ROBUSTUS
E
O. TESSELATUS


B
O. TESSELATUS
METACOXA
C
O. ROBUSTUS


1
2
3
4
5
6
F
O. TESSELATUS


COXITE
STYLUS
D
O. TESSELATUS


4
5
6
G
O. OVALIS

9; TEXAS: Arlington, Avery, Brazos Co., Brownsville, Bryan (Brazos Co.), College Station, Colorado Co., Columbus, Corpus Christi, Daingerfield State Park (Morris Co.), Dallas, Dallas Co., Denton, Devers, Eastland Co., Fedor (Lee Co.), Giddings (Lee Co.), Hildago Co., Houston, Kingsville, Lee Co., Llano (Llano Co.), Peeler, Pleasanton, Riviera Beach (Kleburg Co.), Rock Island, San Jacinto, Sequin, Sulfur Springs, Victoria, Weslaco; March 23 to November 29; ARKANSAS: Almyra, Cove Lake (Logan Co.), Hope, Washington Co., May 28 to July 15; LOUISIANA: Logansport, Vowells Mill; no dates; MISSISSIPPI: Ag. College, A & M College, Gulfport, Lucedale, Waynesboro; April 25 to December 6; ALABAMA: 5 mi. N. E. Eutaw (Greene Co.), Mt. Vernon, Mobile, Tuscaloosa; March 8 to July 9.
Total Specimens Examined: 576.

Omophron dentatus Leconte

Omophron dentatum Leconte, 1852. Ann. Lyceum Nat. Hist., New York, 5: 200–201.
Omophron denatatus Leconte. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 406–407.

HOLOTYPE: Sex (?), repository, Museum of Comparative Zoology, Harvard College. *Type locality:* San Diego, California.

DESCRIPTION

MALES: Total length 5.4–6.6 mm. (M 6.1 mm.), width 3.4–4.3 mm. (M 3.9 mm.), l/w ratio 1.51–1.75 (M 1.59).

FEMALES: Total length 6.1–7.1 mm. (M 6.7 mm.), width 3.7–4.6 mm. (M 4.3 mm.), l/w ratio 1.51–1.64 (M 1.57).

Head: Pale area on front weakly M-shaped (Fig. 2-C); dorsal surface finely punctured; with 4 gular setae (as in Fig. 2-K).

Thorax: Pronotum with an irregular transverse band which extends almost to lateral margins and which possesses 2 or 3 processes extending to posterior margin (as in Fig. 2-I); dorsal surface finely punctured; disc of metasternum sparsely punctured.

Thoracic appendages: Elytron 15-striate, striae moderately impressed, punctures fine, becoming obliterated on apical third dark and pale areas about equal; middle and apical fasciae of elytra usually not joined (Fig. 3-F). Mesocoxae and metacoxae each with 2 setae (as in Fig. 4-B, E).

Abdomen: A pair of ambulatorial setae on 5th sternite (as in Fig. 4-G). Male genitalia with sides of aedeagus parallel, apex broadly truncate (Fig. 5-L).

COMMENTS: This species is one of our larger and more elongate representatives of the group. The maculation of the dorsal surface is a bright metallic green. The ventral surface is blackish in contrast to the legs which are pale testaceous. Distinguishing characters for *dentatus* are: 15-striate elytron, pronotum with an irregular transverse band, light area on front weakly M-shaped, 3 transverse bands of the elytra most often separate (i.e. not joined), and structure of the ♂ genitalia.

The known distribution includes southern California and Arizona. (A single ♀ specimen has been seen from Texas). The northern limit of its range in California appears to be slightly north of San Francisco at a latitude of ca. 39° N. In this region it contacts *solidus*, a northern species which is very closely related. There is no apparent zone of intergradation so that reproductive isolation seems to exist between these two forms.

SPECIMENS EXAMINED: CALIFORNIA: Azuza, Berkeley, Burbank, B. Lake, Camp Greeley (Fresno Co.), Camp Pendleton (Oceanside), Carmel (Monterey Co.), Coarsegold (Madera Co.), Colton, Corona (Santa Ana River), Daggett, Danville, Dimas, Fairfax (Marin Co.), Felton, Fillmore, Fish Camp (Mariposa Co.), Havilah (Kern Co.), Idyllwild (San Jacinto Mts.), Ingleside, Isabella (Kern Co.), Kaweah (Tulare Co.), Lebec, Lone Pine (Inyo Co.), Los Angeles, Los Angcles Co., Los Cerritos, Marin Co., Marsh Creek (Contra Costa Co.), Merced Lake, Miami Ranger Station (Mariposa Co.), Mokelumne Hill (Calaveras Co.), Monterey Co., Niles, Northfork, Oakhurst (Madera Co.), Oceanside, Olancha (Inyo Co.), Owens Lake, Owens Valley, Pasadena, Paso Robles, Pico, Pine Canyon (Mt. Diablo), Pinon Flat (San Jacinto Mts.), Point Reyes, Pomona, Poway (San Diego Co.), Rivera, Riverside, Salina River, San Bernardino Co., San Diego, San Diego Co., San Francisco, San Francisco Co., San Joaquin Exp. R. (Madera Co.), San Mateo Co., Santa Ana, Santa Ana River, Santa Margerita, Santa Monica, Santa Rosa (Sonoma Co.), Santa Rosa Island, Sobre Vista (Sonoma Co.), Stone Cn. (Monterey Co.), Sugar Pine (Madera Co.), Tejon Pass, Tulare Co., Walnut Creek, Whitewater Cn. (Riverside Co.), Yermo; February 16 to November 24; ARIZONA: No locality or date; TEXAS: No locality or date.
Total Specimens Examined: 440.

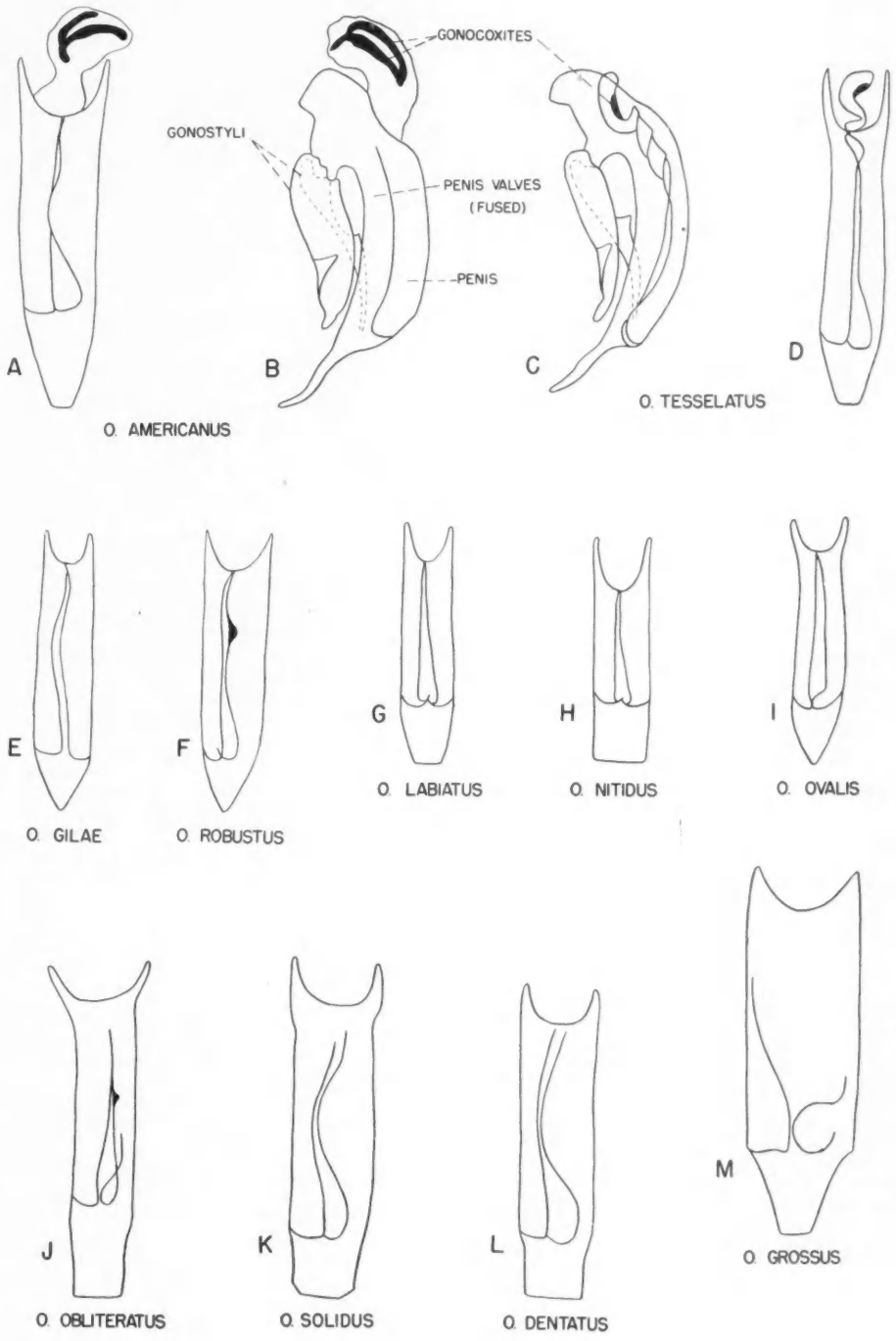
Omophron tesselatus Say

Omophron tesselatus Say, 1823. Jour. Acad. Nat. Sci. Philadelphia, 3: 152.
Omophron lecontei Dejean, 1831. Spec. Gen. Col., 5: 582.
Omophron tesselatum ellipticum Casey, 1909. Canad. Ent., 41: 276.

EXPLANATION OF FIGURE V

Male Genitalia

A. Aedeagus of *O. americanus*, dorsal aspect. B. Aedeagus of *americanus*, dorsal-lateral aspect. C. Aedeagus of *tesselatus*, dorsal-lateral aspect. D Aedeagus of *tesselatus*, dorsal aspect. E. Aedeagus of *gilae*, dorsal aspect. F. Aedeagus of *robustus*, dorsal aspect, showing knob-like structure on ejaculatory duct of penis. G. Aedeagus of *labiatus*, dorsal aspect. H. Aedeagus of *nitidus*, dorsal aspect. I. Aedeagus of *ovalis*, dorsal aspect. J. Aedeagus of *obliteratus* showing knob-like structure on ejaculatory duct of penis. K. Aedeagus of *solidus*, dorsal aspect. L. Aedeagus of *dentatus*, dorsal aspect. M. Aedeagus of *grossus*, dorsal aspect.


GONOCOXITES
GONOSTYLI
PENIS VALVES
(FUSED)
PENIS
A
B
C
D
O. AMERICANUS
O. TESSELATUS
E
F
G
H
I
O. GILAE
O. ROBUSTUS
O. LABIATUS
O. NITIDUS
O. OVALIS
J
K
L
M
O. OBLITERATUS
O. SOLIDUS
O. DENTATUS
O. GROSSUS

HOLOTYPE: Sex (?), repository unknown to authors.[5] *Type locality:* Elk Horn Creek (Missouri).

DESCRIPTION

MALES: Total length 5.4–6.5 mm. (M 6.1 mm.) (S.D. 0.49), width 3.7–4.3 mm. (M 4.1 mm.) (S.D. 0.36), l/w ratio 1.44–1.56 (M 1.50).

FEMALES: Total length 6.0–7.0 mm. (M 6.5 mm.) (S.D. 0.55), width 4.0–4.7 mm. (M 4.4 mm.) (S.D. 0.36), l/w ratio 1.45–1.54 (M 1.49).

Head: Pale area on front M-shaped (Fig. 2-D); dorsal surface with many punctures; with 4 gular setae (Fig. 2-K).

Thorax: Pronotum with a spot on median third (Fig. 2-J); punctures on dorsal surface most prevalent on anterior and posterior margins; disc of metasternum (Fig. 4-E) usually impunctate.

Thoracic appendages: Elytron 15-striate, striae moderately impressed, punctures rather coarse but close set; dark and pale areas about equal (Fig. 3-B). Mesocoxae and metacoxae each with 2 setae (Fig. 4-B, E).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (Fig. 4-F). Male genitalia with sides of aedeagus gradually tapered toward apex, apex rounded, gonocoxites apparently fused (Figs. 5-C, D).

COMMENTS: *O. tesselatus* is morphologically very similar to *grossus* but can be readily distinguished from the latter by its much smaller size. The maculation of the dorsal surface is usually bright metallic green in color, and is generally more extensive than that of *grossus*. Diagnostic characters for this species are: 15 striate elytron, light area on front M-shaped and structure of the ♂ genitalia.

This is one of our most widespread species. Its known geographical range includes Prince Edward Island, New Brunswick, Maine, Quebec, Massachusetts, New York, New Jersey, Pennsylvania, Virginia, Kentucky, Ohio, Indiana, Michigan, Ontario, Manitoba, Saskatchewan, Alberta, North Dakota, Minnesota, Wisconsin, Illinois, Iowa, Nebraska, Kansas, Missouri, Texas, Colorado, New Mexico, Arizona and California.

SPECIMENS EXAMINED: *Holotype: O. tesselatum ellipticum* Casey, ♀, Rhode Island. Repository, U. S. National Museum, Type No. 48080.

ALBERTA: Chappell Lake, Chappice Lake, Medicine Hat; April 15 to August 29; ARIZONA: Yuma; no date; CALIFORNIA: Sunol; May 26; COLORADO: Antonito, Rocky Ford; June 28 to July 28; ILLINOIS: Algonquin, Devil's Neck (Havana), Galesburg, Rock Island, Urbana, Wolf Lake (Chicago); June 22 to September 10; INDIANA: Lafayette, Lawrence Co., Orange Co., Tippecanoe Co., Warsaw; April 28 to July 6; IOWA: Ames, Elma, Fairfax, Iowa City, Ledges State Park, Lost Island Lake (Palo Alto Co.); April 12 to September; KANSAS: Blackjack Creek (Pottawatomie Co.), Medora, Pottawatomie Co., Reno Co., Riley Co., Salt Flats area (Stafford Co.), Sedgwick Co., Superior, Sylvia, Wallace Co., W. Kansas; May 25 to August 16; KENTUCKY: No locality or date; MAINE: Peru; no date; MANITOBA: Aweme, Treesbank, Victoria Beach; April 20 to September 28; MASSACHUSETTS: Natick, White Island Pond (Wareham); April 26 to September 15; MICHIGAN: Black Lake (Cheboygan Co.), Cheboygan Co., Douglas Lake, Washtenaw Co., May 31 to August 10; MINNESOTA: Afton, Baudette, Cass Co., Chisago Co., Crookston, Emily Lake (Lesueur Co.), Gorman Lake (St. Peter), Hennepin Co. (near Shakopee), Itasca Park, Jordan, Lake Pepin, Mille Lacs Lake (near Garrison), Minneapolis, Olmsted Co., Ottawa, Pelican Lake (Nisswa), Pine City, Plummer, St. Paul, Traverse Co., Willmar; April 17 to September 19; MISSOURI: Clarksville, Columbia, Pulaski Co.; June 15 to October 11; NEBRASKA: Atkinson, Carns, Clear Lake (Cherry Co.), Imperial, Lincoln, Mullen, St. Paul, Scotts Bluff, So. Sioux City, Stewart, West Point; June 11 to August 7; NEW JERSEY: Emerson, Mt. View; August 11 to October 11; NEW BRUNSWICK: Penobsquis; July 27; NEW MEXICO: Rio Grande River (near Taos); June 17; NEW YORK: Canton, C. I., Ithaca, L. I., Montauk (L.I.), New York City, Upper Saranac; June 5 to July 8; NORTH DAKOTA: Fish Lake (Turtle Mts.), Jarves Lake (Turtle Mts.), Mission Lake (near Devil's Lake); July 4–19; OHIO: Cincinnati, (6 specimens, no locality or date); June; ONTARIO: Constance Bay, East Ontario, Prince Edward Co., Thunder Bay (Soc des Mille Lacs), Toronto; May 18 to August 13; PENNSYLVANIA: Easton, Harrisburg, Lancaster, Lemoyne, New Cumberland; May 12 to July 5; PRINCE EDWARD ISLAND: Brackley Beach (Canadian National Park); July 2–3; QUEBEC: Lanoraie, St. Jean; April 3 to June 16; SASKATCHEWAN: Attons Lake (Cut Knife), Meeting Lake; June 12 to July 4; TEXAS: No locality or date; VIRGINIA: Vienna; no date; WISCONSIN: Gordon Nursery (Douglas Co.), Griffith St. Nursery (Wood Co.), Kinikinick River (So. Lac St. Croix), lac du Flambeau, Nekoosa (Wood Co.), Oconomowoc, Trout Lake, Waushara Co.; May 23 to August 7.

Total Specimens Examined: 630.

Omophron grossus Casey

Omophron grossum Casey, 1909. Canad. Ent., 41: 275–277.

Omophron grossus Casey, Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 407.

HOLOTYPE: Sex ♂, repository, U. S. National Museum, Type No. 48089, examined. *Type locality:* Texas.

[5]The type specimen was apparently lost with the rest of Say's collection.

DESCRIPTION

MALES: Total length 7.7–8.5 mm. (M 8.2 mm.) (S.D. 0.51), width 5.2–5.8 mm. (M 5.5 mm.)(S.D. 0.30), 1/w ratio 1.46–1.56 (M 1.50).

FEMALES: Total length 8.0–9.1 mm. (M 8.5 mm.) (S.D. 0.50), width 5.4–6.0 mm. (M 5.7 mm.) (S.D. 0.35), 1/w ratio 1.43–1.55 (M 1.50).

Head: Frontal area of head entirely pale (Fig. 2-A); punctures on dorsal surface fine and sparse; with four gular setae (as in Fig. 2-K).

Thorax: Pronotum with spot on median third (as in Fig. 2-J); dorsal surface finely and sparsely punctured; disc of metasternum sparsely punctured.

Thoracic Appendages: Elytron 15-striate, striae moderately impressed, punctures fine and close set; maculation very sparse, exceeded in area by pale ground color (Fig. 3-A). Mesocoxae and metacoxae each with 2 setae (as in Fig. 4-B, E).

Abdomen: Paired ambulatorial setae lacking on 5th sternite (Fig. 4-F). Male genitalia with aedeagus broad, sides parallel but strongly convergent near apex, apex narrowly truncate (Fig. 5-M).

COMMENTS: This species is the largest representative of the genus in North America. The only other species that it could possibly be confused with is *tesselatus* which is very similar morphologically. However, the difference in size is so great that there need not be any question of identity. The maculation of the dorsal surface is metallic green in color and greatly reduced. The transverse bands of the elytra are broken into small irregular spots. *Grossus* is best distinguished from the other species by its great size, 15 striate elytron, pale marking on frontal area, and structure of the ♂ genitalia. The distribution of *grossus* appears to be limited to the south central states. It has been recorded for Iowa, Kansas, Missouri, Arkansas, Oklahoma, and Texas.

SPECIMENS EXAMINED: IOWA: Ames (no date), Iowa City; July; KANSAS: Douglas Co., Garden City, Graham Co., Pottawatomie Co., Riley Co., Sedgwick Co., Topeka; June 8 to AUGUST 16; MISSOURI: Pikes Peak (Pulaski Co.); August 15–19; ARKANSAS: Hope; June 14; OKLAHOMA: Jackson Co.; June 18; TEXAS: Morris Co.; July 3.

Total Specimens Examined: 40.

Omophron solidus Casey

Omophron solidum Casey, 1897. Ann. New York Acad. Sci., 9: 301, 303.

Omophron dentatus, var. *solidus* Casey. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 407.

Omophron lawrencei Hatch, 1953. Part I. Beetles of the Pacific Northwest, p. 69, Univ. of Wash. Press, Seattle. NEW SYNONYMY.

HOLOTYPE: Sex ♂, repository U. S. National Museum, Type No. 48083, examined. *Type locality:* California (Marin to Humboldt Cos.).

DESCRIPTION

MALES: Total length 5.8–7.3 mm. (M 6.7 mm.), width 3.5–4.7 mm. (M 4.2 mm.), 1/w ratio 1.52–1.66 (M 1.59).

FEMALES: Total length 6.5–7.8 mm. (M 7.2 mm.), width 4.0–4.9 mm. (M 4.4 mm.), 1/w ratio 1.59–1.74 (M 1.63).

Head: Pale area on front V-shaped (Fig. 2-B); dorsal surface finely punctured; with 4 gular setae (as in Fig. 2-K).

Thorax: Pronotum with an irregular transverse band which extends almost to lateral margins and which possesses 2 or 3 processes extending to posterior margin (Fig. 2-I); dorsal surface finely punctured; disc of metasternum with few, if any, punctures.

Thoracic appendages: Elytron 15-striate, striae deeply impressed, punctures fine, becoming obliterated on apical third, dark areas usually predominating; middle and apical fasciae of the maculation usually joined in several places giving the elytra a mottled appearance (Fig. 3-C). Mesocoxae and metacoxae each with 2 setae (as in Figs. 4-B, E).

Abdomen: A pair of ambulatorial setae on 5th sternite (as in Fig. 4-G). Male genitalia with sides of aedaegus parallel, becoming angular at apex, apex truncate (Fig. 5-K).

COMMENTS: *O. solidus* is second only to *grossus* in size. The maculation of the dorsal surface ranges in color from a dull black to a bright metallic green. The ventral surface is blackish in contrast to the legs which are pale testaceous. Distinguishing characters of this species are: 15-striate elytron, pronotum with an irregular transverse band, light area on front V-shaped, 3 transverse bands of elytra most often joined in several places giving a mottled appearance; and structure of the ♂ genitalia.

The known distribution includes Oregon and northern California. The southern limit of its range in California appears to be at a latitude of ca. 39° N. Here it contacts *dentatus*, a very closely related but distinct species.

SPECIMENS EXAMINED: *Paratypes: O. solidum* Casey, 4 ♂♂, 5 ♀♀, California. Deposited with holotype. *Paratype: O. lawrencei* Hatch, ♂, Medford, Oregon; June 17, 1935. Deposited with holotype in the Dept. of Zoology, University of Washington.

OREGON: Cow Creek, Medford; July 13–24; CALIFORNIA: Blocksburg, Castella, Duncan Mills (Sonoma Co.), Dyerville (Humboldt Co.), Eldorado Co., Ft. Seward, Guerneville (Sonoma Co.), Hamburg (Siskiyou Co.), Hickey Grove (Mendocino Co.), Humboldt Co., Mesa Grande (Sonoma Co.), Philo (Mendocino Co.), Russian River, Shasta Co., Sobre Vista (Sonoma Co.), Vina (Tehama Co.), Weaverville (Trinity Co.), Willow Creek (Humboldt Co.); May 15 to September 27.

Total Specimens examined: 145.

Omophron ovalis Horn

Omophron ovale Horn, 1870. Trans. Amer. Ent. Soc., 3: 75.
Omophron concinnum Casey, 1897. Ann. New York Acad. Sci., 9: 301, 303.
Omophron gemma Casey, 1897. Ann. New York Acad. Sci., 9: 301, 304.
Omophron frater Casey, 1913. Mem. Col., IV: 41.
Omophron ovalis Horn. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 411.

HOLOTYPE: Sex ♂, repository, The Academy of Natural Sciences of Philadelphia, Type No. 1010, examined. *Type locality:* Ft. Crook, California.

DESCRIPTION

MALES: Total length 4.8–5.8 mm. (M 5.3 mm.), width 3.0–3.8 mm. (M 3.4 mm.), 1/w ratio 1.51–1.68 (M 1.58).

FEMALES: Total length 5.3–6.6 mm. (M 5.9 mm.), width 3.4–4.1 mm. (M 3.7 mm.), 1/w ratio 1.54–1.66 (M 1.60).

Head: Pale area on front V-shaped (as in Fig. 2-B); dorsal surface very finely punctured; with 2 gular setae (Fig. 2-L).

Thorax: Pronotum predominantly dark with pale area on lateral margins irregular in outline (Fig. 2-G); dorsal surface very finely punctured; disc of metasternum impunctate.

Thoracic appendages: Elytron 15-striate, striae moderately impressed, punctures fine and close set; dark areas usually predominating. Mesocoxa with only 1 seta (Fig. 4-A), metacoxa with 2 setae (Fig. 4-B).

Abdomen: Usually with a pair of ambulatorial setae on 5th sternite, often very fine and difficult to see (Fig. 4-G). Apex of aedeagus acute (Fig. 5-I).

COMMENTS: *O. ovalis* is one of the smallest and most elongate species. The maculation of the dorsal surface varies, ranging from a dull dark brown to a bright metallic green. This little beetle most closely resembles *americanus* but is more elongate. Characters which readily distinguish it are: 15 striate elytron, pale area on front V-shaped, 2 gular setae, disc of metasternum impunctate, only one seta on mesocoxa, and structure of the ♂ genitalia.

The known range of this Pacific Northwest species includes British Columbia, Alberta, Washington, Montana, Wyoming, Idaho, Oregon and northern California.

SPECIMENS EXAMINED: *Holotype: O. concinnum* Casey, ♂; Siskiyou Co., California. Repository, U. S. National Museum, Type No. 48084. *Holotype: O. gemma* Casey, ♂, Eel River (Humboldt Co.), California. Repository, U. S. National Museum, Type No. 48081. Also 5 paratypes (1 ♀, 4 ♂♂) labelled California. Deposited with holotype. *Holotype: O. frater* Casey, ♂; California. Repository, U. S. National Museum, Type No. 48082.

BRITISH COLUMBIA: Creston, Fernie, Keremeos, Oliver, Osoyoos, Quamichan Lake (Vancouver Island), Salmon Arm, Tofino, Trinity Valley; April 12 to September 20; ALBERTA: Happy Valley, Lundbreck, Peigan Indian Reservation, Pincher, Pincher Creek; May 22 to July 12; WASHINGTON: Almota, Cedar Mt. (King Co.), Colfax, College Place, Coppei Blue Mts. [sic], Coppei Creek, Dayton, Easton, Ellensburg, Forks (Callam Co.), Klickitat (Glenwood), Kooskooskie, L. McElroy (Paha), Lake Paha, Maloney's Grove (North Bend), Mill Creek (Walla Walla), Moses Lake, Pullman, Ritzville, Sanpoil (Keller), Snake River (Asotin), Spokane, Sprague, Sumner, Toppenish, Walla Walla, Wallula, W. T.; April 13 to September 5; MONTANA: Flathead Co.; Livingston; July 14–15; WYOMING: Green River; July 2; IDAHO: Coeur d'Alene Lake, Juliaetta, Moscow Mt., Sagle (Bonner Co.), Sandpoint, Webb (Nez Perce Co.); April 1 to July 17; OREGON: Corvallis, Dalles, Dilley, Eugene, Gaston, Hood River, McMinnville, Medford, Newport, Pacific City, Portland, Prospect (Jackson Co.), Richland (Baker Co.), Spring Creek (Jefferson Co.), Sucker Creek Canyon (Malheur Co.), Taft, Tygh Valley, Union Creek (Jackson Co.), Vida (Lane Co.), Waldport, Walterville, Woods; April 11 to September 8; CALIFORNIA: Castella, Castle Crag, Cole, Hamburg (Siskiyou Co.), Humboldt Co., Lake Tahoe (Lake Forest), Mohawk, Shasta Co., Siskiyou Co., Trinidad, Trinity Co., Yreka; May 24 to September 22.

Total Specimens Examined: 489.

Omophron americanus Dejean

Omophron americanum Dejean, 1831. Spec. Gen. Col., V, p. 583.
Omphron saii Kirby (sic), 1837. Fauna Bor.-Amer., IV, p. 65.
Omophron lacustre Casey, 1897. Ann. New York Acad. Sci., 9: 301.
Omophron texanum Casey, 1897. Ann. New York Acad. Sci., 9: 301, 302.
Omophron fontinale Casey, 1913. Mem. Col., IV, p. 41.
Omophron iridescens Casey, 1913. Mem. Col., IV, p. 41.
Omophron lengi Casey, 1920. Mem. Col., IX, p. 135. NEW SYNONYMY.
Omophron illustre Casey, 1920. Mem. Col., IX, p. 136. NEW SYNONYMY.
Omophron americanus Dejean. Csiki, 1927. Carabidae I. Col. Cat.—Junk, 1(92): 406.
Homophron tanneri Chandler, 1941. Great Basin Nat., 2: 99–100. NEW SYNONYMY.
Homophron tanneri proximum Chandler, 1941. Great Basin Nat., 2: 99, 101–102. NEW SYNONYMY.
[?] *O. labiatum*, Emmons, 1854 (not Fabricius). Nat. Hist., New York, Agr. Rpt., 5: 52, pl xx, fig. 11.
[?] *O. labiatum* var. *tesselatus*, Emmons, 1854 (not Say). Nat. Hist., New York, Agr. Rpt., 5: 52, fig. 12.

HOLOTYPE: Sex (?), repository unknown to authors. *Type locality:* Northern North America.

DESCRIPTION

MALES: Total length 5.1–6.4 mm. (M 5.7 mm.), width 3.4–4.4 mm. (M 3.9 mm.), 1/w ratio 1.41–1.59 (M 1.48).

FEMALES: Total length 5.2–7.0 mm. (M 6.0 mm.), width 3.3–4.7 mm. (M 4.1 mm.), l/w ratio 1.35–1.60 (M 1.49).

Head: Pale area on front V-shaped (Fig. 2-B); dorsal surface coarsely punctured; with 4 gular setae (Fig. 2-K).

Thorax: Pronotum predominantly dark with pale area on lateral margins irregular in outline (Fig. 2-G); dorsal surface coarsely punctured; disc of metasternum punctate.

Thoracic appendages: Elytron 15-striate, striae usually very deep with convex intervals, punctures close set, becoming indistinct near apex; maculation varies in extent but most often exceeds pale ground color in area (Figs. 3-D, E). Mesocoxae and metacoxae each with 2 setae (Figs. 4-B, E).

Abdomen: Paired ambulatorial setae usually lacking on 5th sternite (Fig. 4-F). Gonocoxites of ♂ genitalia elongate and apparently not fused; aedeagus with sides gradually tapering toward apex; apex narrow and somewhat rounded (Figs. 5-A, B).

COMMENTS: *O. americanus* is our most common North American species and probably the most widespread. Based on structure of the ♂ genitalia, and distribution, it is most closely related to *tesselatus*, yet is readily distinguished by its marking and coloration. It could be confused with *ovalis* (in the west) owing to its dark coloration, but it is generally larger and more robust. Superficially, it resembles *obliteratus* (in the southwest), but is distinguished from that species by its 15-striate elytron. Great variation in the elytral marking and coloration, along with its wide geographical range, has caused past workers to split this species excessively. The variation is so great that it is virtually impossible to generalize with regard to marking. Individuals in the northeastern United States tend to be generally darker, i.e. the ground color as well as the maculation of the elytra, is darker. Because the maculation is more extensive (Fig. 3-D), the fasciae are often joined in several places; also, the elytral striae tend to be deeper. Individuals in the southwestern and western extremes of the range tend to have the elytral maculation less extensive (Fig. 3-E), often exceeded in area by a lighter ground color, and the striae are less impressed. For the species in general, the maculation of both pronotum and elytra is tinged with metallic green in varying degrees.

The diagnostic characters for this species are: 15-striate elytron, pale area on front V-shaped, pronotum predominantly dark with pale areas on lateral margins irregular in outline, 2 setae on mesocoxa, disc of metasternum punctate, and structure of the ♂ genitalia.

The species illustrated and described by Emmons as *labiatum* and *labiatum* var. *tesselatus* do not resemble these species at all but are much more like *americanus*. The descriptions are not adequate for positive recognition. We consider these to most resemble *americanus*, the most common species found in New York.

O. americanus is recorded for Prince Edward Island, Nova Scotia, New Brunswick, Quebec, Maine, New Hamsphire, Massachusetts, Connecticut, New York, New Jersey, Pennsylvania, Delaware, Maryland, District of Columbia, Virginia, North Carolina, South Carolina, Florida, Alabama, Mississippi, Louisiana, Arkansas, Tennessee, Missouri, Kentucky, Ohio, Indiana, Illinois, Wisconsin, Michigan, Ontario, Manitoba, North Dakota, Minnesota, Iowa, Nebraska, Kansas, Oklahoma, Texas, Mexico, New Mexico, Arizona, Colorado, Utah, Idaho, Alberta, Saskatchewan and Northwest Territories.

SPECIMENS EXAMINED: *Holotype: O. lacustre* Casey, ♀; Lake Superior, Bayfield, Wisconsin. Repository, U. S. National Museum, Type No. 48086. *Holotype: O. texanum* Casey, ♀; Colorado River, Austin, Texas; June; Repository, U. S. National Museum, Type No. 48088. *Holotype: O. fontinale* Casey, ♂; Jemez Springs, New Mexico. Repository, U. S. National Museum, Type No. 48090. Also 1 ♂ paratype labelled New Mexico, deposited with holotype. *Holotype: O. iridescens* Casey, ♂; Vicksburg, Mississippi. Repository, U. S. National Museum, Type No. 48079. *Holotype: O. lengi* Casey, ♂; South Carolina. Repository, U. S. National Museum, Type No. 48085. Also 1 ♂ paratype labelled South Carolina, deposited at California Academy of Sciences. *Holotype: O. illustre* Casey, ♀; Vineyard, Utah. Repository, U. S. National Museum, Type No. 48094. *Paratypes: Homophron tanneri* Chandler, 2 ♀♀; Moab, Utah. Deposited at California Academy of Sciences. *Paratypes: Homophron tanneri proximum* Chandler, 1 ♂, 1 ♀; Box Canyon near junction of Escalante River and Calf Creek, Garfield Co., Utah. Deposited at California Academy of Sciences.

ALABAMA: Barnesville, Tuscaloosa; June 8 to August 23; ALBERTA: Chappice Lake,Medicine Hat; March 31 to September 22; ARIZONA: Botatakin (Navajo Nat'l Mon.), Kayenta (15 mi. NNW); June 24 to July 2; ARKANSAS: Cove Lake (Logan Co.), Hope; June 11–29; COLORADO: (1 specimen, no locality or date), Mineral Springs; August; CONNECTICUT: Cornwall, New Haven; May 9 to August 23; DELAWARE: Dover, Water Gap; July 1 to August 30; DISTRICT OF COLUMBIA: May 2; FLORIDA: Branford; July 31; IDAHO: No. Shore Bear Lake (Bear Lake Co.); July 6; ILLINOIS: Algonquin, Chicago, Galesburg, Havana, Lake Co., No. Illinois, Putnam Co., Salt Fork (Urbana), Watson; April 11 to September 11; INDIANA: Lafayette, Marshall Co., N. Judson, Pine, Putnam Co., Starke Co., Steuben Co., Tippecanoe Co., Vigo Co., Warren Co., Winona Lake; April 21 to August 8; IOWA:

Ames, Elma, Mt. Pleasant; May 1 to September 10; KANSAS: Blackjack Creek (Pottawatomie Co.), Doniphan Co., Douglas Co., Ft. Scott, Franklin Co., Lawrence, McPherson, Mt. Hope, Muncie, Onaga, Pottawatomie Co., Tonganoxie, Riley Co., Topeka, Wallace; April 28 to October 14; KENTUCKY: (1 specimen no locality or date), Henderson; May 15; LOUISIANA: Vernon, Vowell's Mill; no dates; MAINE: Monmouth, Wales; June 21–23; MANITOBA: Aweme, Victoria Beach; July 1 to August 11; MARYLAND: Baltimore, Bladensburg, Cabin John, Catonsville, Montgomery Co., Plummers Island; May 12 to September 21; MASSACHUSETTS: Amherst, Brookline, Chickopee, Framingham, Holliston, Natick, Stoneham, White Island Pond (Wareham); April 21 to September 17; MICHIGAN: Agr. College, Ann Arbor (Washtenaw Co.), Cheboygan Co., Cliff (Keweenaw Co.), Detroit, Dexter, Douglas Lake, Grand Ledge, Grand Rapids, Honey Creek (Washtenaw Co.), Livingston Co., Seneca Lake (Keweenaw Co.), Third Sister Lake (Ann Arbor), Whitefish Point (Lake Superior); April 24 to October 6; MINNESOTA: Afton, Anoka Co., Chisago Co., Coon Creek (Anoka Co.), Crystal Lake, Garrison, Hennepin Co., Itasca Park, Lake Cormorant (Becker Co.), Lake Johanna (Ramsey Co.), Lake Minnetonka, Mille Lacs Lake, Nerstrand Woods (Rice Co.), New Brighton, Olmsted Co., Ottawa, Pelican Lake (Nisswa), Pine Co., St. Paul, Vineland; April 17 to October 14; MISSISSIPPI: Waynesboro; July 12; MISSOURI: Bonfils, Columbia; April 14 to October 12; NEBRASKA: Alliance, Clear Lake (Cherry Co.), Halsey, Holt Co., So. Sioux City, Trout Lake (Cherry Co.), Waverly, West Point; April 2 to August 6; NEW BRUNSWICK. Kennebecasis River (Penobsquis), Penobsquis; July 26–27; NEW HAMPSHIRE: Antrim, Franconia, North Conway, Manchester; June 1 to September 20; NEW JERSEY: Berkeley Heights, Boonton (Morris Co.), Clementon, Emerson, Hamden, Irvington, Morristown, Oradell, Phillipsburg, Swartswood Lake, "Upper" Montclair, Westwood (Bergen Co.); April 6 to August 31; NEW MEXICO: Albuquerque, Grant Co., Jemez Mts., Rio Grande River (near Taos), Santa Fe, Santa Fe Canyon; June 4 to August 4; NEW YORK: Barre, Bay Shore (L.I.), Buffalo, Canton, Fair Haven, Florida, Great Hills (S.I.), Inlet Valley (Ithaca), Ithaca, Lancaster, Litchfield Creek, Lockport, Long Island, New York City, Nyack (Rockland Co.), Orangeburg, Peekskill, Pike, Pine Island, Skinnecock Hills (L.J.), Southold (L.I.), Staten Island, W. Hebron, White Plains (Westchester Co.), Willow Brook, Wyandanch (L. I.); April 4 to October 16; NORTH CAROLINA: Swannanoa Valley; May 23; NORTH DAKOTA: N. Odessa Pond (Devil's Lake), Williston; July 4–9; NORTHWEST TERRITORIES: No locality or date; NOVA SCOTIA: Truro, West River; May 18 to August 19; OHIO: Athens Co., Cedar Point, City Park (Athens), Columbus; June 6 to October 14; OKLAHOMA: Beckham Co., Greer Co., Jackson Co., Pottawatomie Co., Texas Co., Wichita Nat'l Forest; June 7 to August 9; ONTARIO: Colony, E. Ontario, Normandale, Osgoode, Ottawa, Sand Pits (Ottawa); May 30 to July 9; PENNSYLVANIA: Aspinwall, Belfast, Easton, Fernwood, Germantown, Philadelphia, Phillipsburg, Wyoming; April 18 to August 27; PRINCE EDWARD ISLAND: Brackley Beach (Canadian National Park); July 2; QUEBEC: Brome, Ft. Coulonge, Ile Jesus, Ille d'Orleans, Lanoraie, Montreal, St. Jean; May 24 to October 11; SASKATCHEWAN: Attons Lake (Cut Knife); June 15 to September 4; SOUTH CAROLINA: Clemson College; April 26 to June 2; TENNESSEE: Memphis; September 27; TEXAS: Austin, Bandera (15 mi. so.), Brownsville (Esper. Ranch), Castolon, College Station, Colletto Creek (Victoria), Colorado Co., Columbus, Corpus Christi, Dallas, Dimmit Co., Eastland Co., Florence, Llano (Llano Co.), Rock Island, Round Mt., Shovel Mt., Sweetwater, Trinity River (8 mi. east of Dallas, Tarant Co.), Victoria, Weslace; March 3 to October 11; UTAH: Moab, Utah Lake (East Side); no dates; VIRGINIA: Black Pond, Buffalo Creek, Fredricksburg, Nelson Co.; May 1 to October 7; WISCONSIN: Madison (Dane Co.), Trout Lake, Waushara Co.; May 4 to June 30.

MEXICO: COAHUILA: Saltillo; May 23; VERA CRUZ: No locality or date.

Total Specimens Examined: 1008.

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SOME STUDIES ON THE INFLUENCE OF LIGHT ON THE MATING ACTIVITY OF ANOPHELES QUADRIMACULATUS SAY[1]

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During the course of previously described studies on the laboratory biology of *Anopheles quadrimaculatus* Say (Terzian & Stahler, 1954), it became apparent that light was an important ecological factor in determining the mating behavior of this mosquito. Accordingly, studies were undertaken to define certain aspects of the manner in which light affects the sexual activity of *A. quadrimaculatus*. This paper reports the results of investigations on the effects of natural and artificial illumination, and of changes in light intensity, on diurnal mating rhythm and the consequent rates of insemination.

[1]The opinions or assertions contained herein are the private ones of the writers and are not to be construed as official or reflecting the views of the Navy Department or the naval service at large.

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MATERIALS AND METHODS

The experiments were performed in three connecting laboratory rooms which were maintained at a constant temperature of 80° F. and a relative humidity of 70 to 75 percent. In one room, in which stock colonies of *A. quadrimaculatus* were kept, and from which the eggs to be used for experimental purposes were collected, the only source of light was that provided by overhead daylight fluorescent lamps. The maximum intensity of light within the experimental cylinders maintained in this room was 4.0 ± 0.5 foot candles, as measured by means of a Weston microammeter and a Weston photronic cell.

In another room, in which the larval colonies were reared and the pupae collected, the sources of illumination were sunlight transmitted through windows with a southeastern exposure and over-

head daylight fluorescent lamps which remained lighted during working hours. The maximum intensity of light recorded in the experimental cylinders in this room was 30 ft. c. In the afternoon the decline in light intensity began at about 3:00 p.m. in winter and at about 5:00 p.m. in summer, and is illustrated by the following readings made of the intensity of light within the experimental cylinders at various times during 2 days:

October 22		May 13	
Time (p.m.)	Light Intensity (ft. c.)	Time (p.m.)	Light Intensity (ft. c.)
1:30	26.0	1:00	26.0
3:00	19.0	5:00	20.0
4:00	10.0	6:15	11.5
4:50	3.5	6:55	4.2
5:05	2.0	7:45	0.8
5:20 (sunset)	0.5	8:12 (sunset)	0.5
5:35	0.05	8:31	0.01

In numerous observations over an entire year, the maximum intensity of light within the cylinders at sunset varied from 0.3 to 0.5 ft. c. During the course of these experiments there was no artificial illumination in this room from about 3:30 p.m. until about 8:00 a.m. on the following morning, so that the only light around sunset and sunrise was that derived from solar radiation. Therefore, the major conditions of illumination in this room will be designated as "natural light."

In the third room illumination was provided by a single daylight fluorescent lamp which was controlled by a clock mechanism normally set so that during each 24-hour period there was light only from 5:30 a.m. to 5:30 p.m. However, continuous light or darkness could be maintained throughout the entire course of an experiment, if so desired. The maximum light intensity within the experimental cylinders in this room was 0.1 ft. c.

The strain of *A. quadrimaculatus* used in these experiments was obtained originally from the U. S. Department of Agriculture at Beltsville, Md. in 1945. Stock colonies, housed in wire-mesh cages of the sleeve type (32″ x 25″ x 25″), were routinely provided with cooked raisins and periodically allowed to feed on guinea pigs in order to provide eggs for continuous breeding. Larvae were reared in white enameled photographic pans (20″ x 17″ x 2½″), with each colony consisting of about 250 individuals. The larvae were fed Difco Brain Heart Infusion, brewer's yeast and finely powdered (100 mesh) Purina Laboratory Chow, according to the method described by Heal and Pergin (1945). On this regimen pupation began on the ninth day and ended on the eleventh day.

The mosquitoes to be used for experimental purposes were separated according to sex in the pupal stage and allowed to emerge into glass lantern globes, which were covered at the top with bobbinette and held upright by metal stands. The adult mosquitoes were supplied with a 4 percent sugar solution, which was routinely changed every day.

The population within each experimental cylinder consisted of 20 males and/or 20 females. In all instances, except for those experiments listed in Table VII, the males were withdrawn from the cylinders by means of a sucking tube at the beginning of each period of cohabitation and transferred in this manner to the cylinders containing the female mosquitoes. A dim red safety light was used to assist in the transfer of male mosquitoes whenever cohabitation was to take place in darkness.

Unless otherwise stated, all observations on the rates of insemination were made with 4-day old mosquitoes, since it has been shown that *A. quadrimaculatus* is most active sexually at this age (Terzian and Stahler, 1954). To establish the proportion of inseminated females within an experimental group, all females were dissected and the spermathecae examined for the presence or absence of spermatozoa.

RESULTS

Diurnal mating rhythm. It has been well established that if males and females of *A. quadrimaculatus* are allowed to cohabit for a 24-hour period of day and night, a high rate of insemination will occur during this time (Terzian and Stahler, 1954). The following experiments were designed to determine whether or not mating activity varied during certain periods of the day or night. The salient points of reference for each 24-hour period were considered to be the time of sunrise and of sunset, as indicated by the Washington, D. C. Weather Bureau data.

In these experiments, male and female mosquitoes were paired for various periods of cohabitation, lasting for 4, 8, 12, 16, and 24 hours. In the first experiment, three groups of males were united with three groups of females at 8 a.m. and one of each resultant colony was permitted to remain in cohabitation until 4 p.m., 8 p.m., or 8 a.m. on the following day, respectively. Thus, in the first mating period, the mosquitoes could have been influenced neither by sunrise nor by sunset; during the second they were exposed only to sunset; and in the third they cohabited during both sunset and sunrise. In the next experiment, two groups of males and females were paired at 4 p.m., with one the period of cohabitation was concluded at 8 p.m., so that mating could take place during sunset but not at sunrise, while in the second group cohabitation was not terminated until 8 a.m. on the following day, in order for the sexes to be together during

both dusk and dawn. Finally, two mating periods were selected so as to begin at 8 p.m., with one ending at midnight, in order to determine the nocturnal rate of insemination during that interval of time, and the other terminating at 8 a.m. on the following day, so that sunrise, but not sunset could be an influencing factor during this period of cohabitation. Three series of such experiments were performed, one in February, another in July, and a third in November. In each experiment, two groups were dissected at the conclusion of every period of cohabitation, so that the rates of insemination were determined for 40 female mosquitoes.

The results, as presented in Table I, indicate that mating occurred mainly during the 4-hour interval that included sunset. For example, in the experiment of February 11, the rates of insemination in those groups which had cohabited

TABLE I

THE PROPORTIONS OF INSEMINATED FEMALES OF *Anopheles quadrimaculatus* DURING VARIOUS INTERVALS OF A SINGLE DAY UNDER NATURAL CONDITIONS OF LIGHT AND DARK

PERIOD OF COHABITATION	AVERAGE PERCENT OF INSEMINATED FEMALES IN TWO EXPERIMENTAL GROUPS			
Time	No. of Hrs.	Feb. 11*	Jul. 28†	Nov. 16#
8 a.m. to 4 p.m.	8	0	0	0
8 a.m. to 8 p.m.	12	68	37	63
8 a.m. to 8 a.m.	24	74	35	83
4 p.m. to 8 p.m.	4	65	57	65
4 p.m. to 8 a.m.	16	67	45	70
8 p.m. to 12 p.m.	4	0	0	5
8 p.m. to 8 a.m.	12	20	15	43

*Sunrise 7:05 a.m.; sunset 5:41 p.m.
†Sunrise 5:05 a.m.; sunset 7:15 p.m.
#Sunrise 7:21 a.m.; sunset 4:48 p.m.

at least during the sunset period varied from 65 to 74 percent, while in the remaining groups which had mated during portions of the day and night other than sunset, the rates of insemination ranged from 9 to 20 percent. The same pattern of mating activity was repeated in the other two experiments.

These data indicate further that, although there was some mating at sunrise, it was considerably less than that which occurred during sunset. In those groups exposed only to the influence of sunset, the average rate of insemination was 59 percent, while in groups which cohabited during sunrise, but not during sunset, only 26 percent of the females became inseminated.

It is apparent, too, that there was little or no mating during the day, between sunrise and sunset, or at night, between dusk and dawn. These observations are in general agreement with those of Roth (1948), who found that *A. quadrimaculatus* usually mated at 6 p.m. and 7 a.m. Complete information on nocturnal mating activity is lacking, since no periods of cohabitation were begun immediately after sunset and terminated just before dawn. However, in the experiments in which the period of cohabitation lasted from 8 p.m. to midnight, the sharp decline in rates of insemination would suggest that mating activity could be re-established only after an increase in light intensity.

The data also indicate that, within a single day, the number of daylight hours prior to sunset in which the mosquitoes have been cohabiting does not materially affect the rate of insemination at sunset. Thus, in a group which had been paired 9 hours before sunset, the proportion of inseminated females after sunset was 63 percent, whereas in another group in the same experiment in which the mosquitoes had been paired only 1 hour before sunset on the same day, 65 percent of the females became inseminated.

The peaks of mating activity during the 24-hour cycle of day and night were the same, namely at sunset, in experiments performed in February, July and November, notwithstanding the nearly 2½ hours difference in sunset time between two of the experiments. This indicates that changes in mating activity are dependent on sunrise and the sunset times rather than on certain chronometric times.

Since the greatest mating activity occurred during the 4-hour period which included sunset, the following experiments were designed to define more accurately the time interval when mating activity was maximal. Accordingly, mosquitoes were paired for 30 to 60 minute periods before, during, and after sunset. The results of these experiments, presented in Table II, indicate, in every case, that throughout the year the peak of mating activity occurred during the half-hour interval which included sunset. Furthermore, there was little or no mating activity in any of the groups until approximately 30 minutes before sunset, and it ceased just as abruptly about 30 minutes following sunset.

The results of the experiment performed on July 1 are of particular interest. On that afternoon the natural light intensity varied considerably, because of occasional thundershowers and an overcast sky, and, as a result, active mating began about 2 hours prior to sunset and continued during 4 consecutive one-hour periods of cohabitation, or about twice as long as in the other experiments listed in Table II. Somewhat similar mating behavior, departing from the usual pattern, was noted on several other dark and cloudy days, indicating again that certain light intensities or changes in light intensity simulating sunset are more important factors in determining mating activity than is some inherent "biological clock."

Thus the type of diurnal mating rhythm dis-

played by this mosquito should be characterized, according to the terminology of Park (1941) as "exogenous," that is, it is a periodic activity in which the pattern is directly induced and controlled by the periodic environmental factors. This is in contrast to the "endogenous" type of rhythmic activity in which the pattern is resident in the animal itself and therefore more deeply seated, as evidenced in the persistent locomotor rhythm in the beetle, *Boletotherus* (Park and Keller, 1932).

Effect of light adaptation on mating at sunset. To furnish additional information as to the extent to which light adaptation before cohabitation influences the typical mating pattern in which insemination occurs mainly at sunset, the mating behaviour of experimental groups which had been adapted to constant artificial light or darkness was compared, during sunset, to groups maintained under natural conditions of light and dark. Thus, one series of experimental groups was maintained for the first 4 post-emergence days in the natural cycle of day and night, a second series was conditioned in a constant artificial light (0.1 ft. c.), a third series was kept for the same length of time in another constant artificial light (4.0 ft. c.), while, finally, a fourth series remained in total darkness. On the fourth post-emergence day starting 1½ hours before sunset, males and females in each experimental group were paired in natural light for six successive half-hour periods of cohabitation.

It is evident from the data in Table III that light adaptation influences the diurnal mating rhythm, since mating activity during the sunset period was modified in groups conditioned in artificial light or in darkness. Whereas 75 percent of the females became inseminated during sunset in an experimental group maintained continually in natural light and dark, only 25 and 37 percent insemination occurred at the same time in the two groups which had been conditioned in artificial light, and 10 percent in the group previously maintained in darkness. Furthermore, the rates of insemination during the sunset period were higher in those groups transferred from 4.0 ft. c. to natural light than in groups taken from 0.1 ft. c., possibly because of the greater change of light intensity which occurred in the former series. The effect of conditioning is even more apparent in the dark-adapted groups where no evident diurnal mating rhythm occurred and the rates of insemination were consistently low over the 3-hour span of cohabitation.

TABLE II

THE RATE OF INSEMINATION IN *A. quadrimaculatus* IN RELATION TO SUNSET IN NATURAL LIGHT

	Period of Cohabitation and Average Rate of Insemination of the Females of Two Experimental Groups						
Date.....	May 13	Jul. 1	Sep. 9	Sep. 11	Oct. 22	Nov. 4	Nov. 11
Sunset Time...	7:12	7:38	6:26	6:23	5:20	5:05	4:58
	1:50–2:50 0	3:15–4:15 0	4:40–5:10 0	2:00–3:00 0	4:20–4:50 0	4:20–4:50 0	3:15–3:45 0
	2:50–3:50 0	4:15–5:15 0	5:10–5:40 0	3:00–4:00 0	4:50–5:50 42	4:50–5:20 35	3:45–4:15 0
	3:50–4:50 0	5:15–6:15 40	5:40–6:10 10	4:00–5:00 0	5:05–5:35 50	5:20–5:50 15	4:15–4:45 0
	4:50–5:50 0	6:15–7:15 25	6:10–6:40 75	5:00–6:00 0	5:20–5:50 38	5:50–6:20 0	4:45–5:15 53
	5:50–6:50 32	7:15–8:15 55	6:40–7:10 61	6:00–7:00 62	5:50–6:20 0		5:15–5:45 33
	6:50–7:50 67	8:15–9:15 20	7:10–7:40 0	7:00–8:00 2			5:45–6:15 0
	7:50–8:50 0	9:15–10:15 5					
	8:50–9:50 0						

It is evident from the data in Table III that light adaptation influences the diurnal mating rhythm, since mating activity during the sunset

Mating in artificial light. Since it had been shown that mating activity at sunset is influenced by light adaptation, the rates of insemination were compared in uniformly-adapted mosquitoes cohabiting in various conditions of illumination. For this purpose, three experimental groups were maintained in the natural cycle of light and dark for the first four post-emergence days and at 15 minutes before sunset on the fourth day they were transferred to and paired in artificial light or darkness and then permitted to cohabit for 30 minutes. For purposes of comparison, another similarly conditioned group was allowed to mate in natural light during sunset. Five such experiments were performed, with results as indicated in Table IV.

The data suggest that mating activity was significantly affected when the mosquitoes were removed from natural light at sunset to artificial light (0.1 or 4.0 ft. c.) or to darkness. Thus, the rates of insemination in those groups transferred to 0.1 ft. c. were only about half as much as in the groups remaining in natural light, while the groups transferred to 4.0 ft. c. or to total darkness showed very little or no mating activity. It would appear, therefore, that the stimulus to mating produced by the appropriate light intensity or by changing the light intensity ceases immediately if the transfer is from an optimal environment to a very unfavorable one, resulting in marked changes in the consequent rates of insemination.

TABLE III

THE RATES OF INSEMINATION OF *A. quadrimaculatus* CONDITIONED FOR 4 DAYS AT VARIOUS LIGHT INTENSITIES AND MATED IN NATURAL LIGHT DURING THE SUNSET PERIOD

Cohabitation Period (Sunset at 7:58 P.M.)	Light Intensities During Adaptation Period			
	Natural Light	0.1 ft. c.	4.0 ft. c.	Darkness
6:15 to 6:45	0	0	0	6
6:45 to 7:15	0	0	5	0
7:15 to 7:45	10	0	32	15
7:45 to 8:15	75	25	37	10
8:15 to 8:45	61	0	0	5
8:45 to 9:15	0	0	0	0

TABLE IV

THE RATES OF INSEMINATION OF *A. quadrimaculatus* CONDITIONED FOR 4 DAYS IN NATURAL LIGHT AND MATED AT DIFFERENT LIGHT INTENSITIES DURING SUNSET PERIOD

Light Intensity During 30 Min. Cohabitation at Sunset Period	Percent Females Inseminated in five Experimental Groups
Natural Light	60
0.1 ft. c.	33
4.0 ft. c.	5
Darkness	0

Mating response to decrease in light intensity. In view of the fact that insemination did not readily occur in artificial light of a constant intensity, it was necessary to assess the mating response to a sudden decrease in artificial light intensity. Thus, on the fourth post-emergence day, a series of experimental groups which had been maintained in a constant artificial light intensity of 4.0 ft. c. since emergence, were transferred simultaneously to another constant artificial light of 0.1 ft. c. intensity. Immediately after the transfer from one environment to the other and at half-hour intervals thereafter, for a period of 10½ hours, groups of males were paired with groups of females and allowed to cohabit for 30 minutes. The consequent rates of insemination, as determined from five series of experiments, are listed in Table V.

These data indicate that, as a result of the transfer from a luminosity of 4.0 ft. c. to one of 0.1 ft. c. and the consequent drop in lightint ensity, there was an immediate mating response. Mating activity decreased steadily, however, with time, as the mosquitoes became conditioned for increasingly longer periods to the lower light intensity. Thus, in mosquitoes paired immediately after transfer, 37 percent of the females became inseminated, but within 1 hour the rate declined to 14 percent, and then continued to decline until at the end of 11 hours following the original transfer, there was no mating activity at all. It would appear as if the change in light intensity from 4.0 to 0.1 ft. c. provides a stimulus for mating but that the response is of no longer duration than 1 hour.

TABLE V

THE EFFECT OF A SUDDEN CHANGE IN ARTIFICIAL LIGHT INTENSITY ON THE RATE OF INSEMINATION OF *A. quadrimaculatus*, MAINTAINED AT 4.0 FT. C., THEN TRANSFERRED TO 0.1 FT. C., AND KEPT AT 0.1 FT. C. FOR VARYING PERIODS OF TIME BEFORE BEING PERMITTED TO MATE

Length of Time at 0.1 ft. c. Before Cohabiting	Average Rates of Insemination in five experimental Groups After 30 Min. Cohabitation at 0.1 ft. c.
0 minutes	37
30 "	24
60 "	14
90 "	9
120 "	12
150 "	8
180 "	13
210 "	5
360 "	5
640 "	0

To determine whether the act of transferring experimental cylinders of mosquitoes from one room to another might have had some mechanical effect on sexual activity, other groups were agitated immediately prior to being paired at the light intensity at which they had been conditioned. Since no insemination occurred in any of these groups, it would seem as if the change in light intensity, rather than mechanical agitation or sudden flight activity, was the significant factor in determining the extent of mating activity.

Since it has not yet been clearly demonstrated whether the stimulus to mating is greater in the

one sex than it is in the other, it was considered of interest to evaluate the mating response of male and female *quadrimaculatus* when exposed to the same change in light intensity as described in the previous experiment. Accordingly, in the first of the following series of experiments, 14 groups of male mosquitoes were emerged into and maintained in an artificial light of 4.0 ft. c. intensity. On the fourth post-emergence day each group of males was transferred to another artificial light of 0.1 ft. c. intensity and immediately thereafter paired with a comparable group of females which had been conditioned concurrently at 0.1 ft. c. In another series of experiments, 14 groups of females were conditioned in the same manner at 4.0 ft. c., the males at 0.1 ft. c., and mating likwise occurred at the lower light intensity. In a third series of 14 groups, males and females were conditioned at 4.0 ft. c. and transferred to 0.1 ft. c. immediately before being paired, so that the stimulus of a decrease in light intensity would act simultaneously on both sexes. In each series, seven groups were allowed to cohabit for 30 minutes and an equal number for 60-minute periods.

TABLE VI

THE EFFECT ON EACH SEX OF A DECREASE IN ARTIFICIAL LIGHT INTENSITY FROM 4.0 TO 0.1 FT. C. IMMEDIATELY PRIOR TO PAIRING ON THE RATE OF INSEMINATION OF *A. quadrimaculatus*

Sex	Period of Cohabitation and Average Rate of Insemination in Seven Experimental Groups	
	30 min.	60 min.
Males	7	27
Females	15	24
Males and Females	23	56

The results of these experiments, presented in Table VI, indicate no significant differences in the rates of insemination after stimulation by a change in light intensity applied to one sex or the other. It is of interest to note, too, that when both sexes were stimulated simultaneously, the mating activity was almost exactly double that when only one sex was stimulated. Since polygamous mating is found in *A. quadrimaculatus*, it might be assumed that the stimulation of both sexes together would result in rates of insemination higher than the sum of those obtained upon the activation of either sex separately. However, it appears not unlikely that polygamous mating does not occur within the first hour of cohabitation under these conditions mainly because this particular stimulus elicits a mating response of only a low order. Further investigation, particularly under circumstances more nearly those of natural sunset, may reveal sexual differences in the response of *A. quadrimaculatus* to gradually decreasing luminous intensities, but these differences were not evident as a result of these studies.

Effect of light during 5 days cohabitation. The studies on the effect of light on mating activity previously described have been mainly limited to 60 minutes or less of continuous cohabitation, with some observations extending up through a 24-hour mating period. The following experiment was designed to evaluate the effect of various conditions of light on the rates of insemination over longer periods of time. Accordingly, 10 groups of males and females were allowed to emerge and remain together for periods ranging from 1 to 5 days in each of the following conditions: natural light and dark, constant artificial light (0.1 and 4.0 ft. c.), and continuous darkness. During each of the first five post-emergence days, the females in two groups for each category were dissected to determine the rates of insemination up to and including that period of cohabitation.

TABLE VII

THE EFFECT OF CONDITIONS OF ILLUMINATION ON THE RATE OF INSEMINATION OF *A. quadrimaculatus* IN CONTINUOUS COHABITATION

Period of Cohabitation (Days)	Average Percent of Insemination in two Groups			
	Natural Light and Dark	Constant Artificial Light		Darkness
		(0.1 ft. c.)	(4.0 ft. c.)	
1	0	0	0	0
2	70	5	0	0
3	72	22	8	0
4	82	45	29	0
5	95	41	43	0

The data, as presented in Table VII, indicate that no mating occurred in any of the groups during the first post-emergence day. On the 2nd day, however, in the groups exposed to natural sunset, the proportions of inseminated females were already near maximal and by the fifth day, 95 percent of the females had mated. The data also indicate that insemination occurs less readily in mosquitoes exposed to a constant artificial light of 0.1 or 4.0 ft. c. intensity, since under these conditions, only about 40% of the females became inseminated by the fifth day. It is further evident that in artificial light, the proportions of inseminated females were lower during each of the first 4 days of cohabitation at the greater intensity (4.0 ft. c.), but that they reached the same level as that in the mosquitoes mating at 0.1 ft. c. on the fifth day. Thus, it is apparent that in *A. quadrimaculatus* mating is not dependent on changes in light intensity and will occur in a constant illumination, with the

degree of activity influenced by the particular light intensity. Finally, the results of this experiment demonstrate that the absence of light completely inhibits successful mating activity, even over a 5-day period of cohabitation.

SUMMARY

It has been established that there is a definite rhythm in the mating activity of *Anopheles quadrimaculatus* during the 24-hour period of day and night. Thus, under natural conditions of light and dark, there was a maximum of mating activity at sunset, some mating at sunrise, but no mating at all between these two periods.

In addition, it was shown that the peaks of mating activity were related to sunrise and sunset times during all seasons of the year, indicating that chronometric times play no important role in the daily mating activity of this mosquito.

Further, it was demonstrated that mating activity at sunset was influenced by previous light adaptation, since the rates of insemination at that period were maximal among mosquitoes which had been conditioned to natural light, less among those which had been adapted to a constant artificial light (0.1 ft. c.), and minimal in those which had been conditioned to darkness.

It was also shown that transferring the mosquitoes to artificial light of too high an intensity (4.0 ft. c.) or to darkness during sunset, when the rates of insemination were usually greatest, interrupted mating activity, indicating that such activity could not be sustained in an unfavorable illumination.

In addition, it was found that a decrease in artificial light intensity from 4.0 to 0.1 ft. c. stimulated mating, although the duration of the response to such stimulus was less than 1 hour. There was no difference in the consequent rates of insemination upon exposing males or females alone to a decreased light intensity.

Finally, it has been shown that, over a 5-day period of continuous cohabitation, the maximal rates of insemination occurred in those groups kept in the natural cycle of day and night, under the influence of changes in light intensity associated with sunrise and sunset. There was less mating activity in groups maintained in a constant artificial light of 0.1 ft. c. and still less for the first 4 days at 4.0 ft. c., but the fact that mating did occur in these environments suggests that changes in light intensity are not prerequisite for mating in *A. quadrimaculatus*. In a constant illumination the intensity of light is probably an important factor in determining the rate of insemination. No mating at all occurred in mosquitoes kept in darkness, even over a 5-day period.

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THE NUMBERS OF EGGS DEVELOPED RELATED TO THE QUANTITIES OF HUMAN BLOOD INGESTED IN AEDES AEGYPTI (L.) (DIPTERA: CULICIDAE)[1]

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The basic needs of bloodsucking arthropods are being investigated as a means toward understanding their behavior in relation to the vertebrate host. The results reported in this article extend our information on the relation between the numbers of eggs that are developed by *Aedes aegypti* (Linnaeus) and the varying quantities of human blood ingested as affected by differences between individual mosquitoes, ages of the mosquitoes at the time of the blood meal, and weights of the mosquitoes before the blood meal.

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Roy (1936) found that *Aedes aegypti* fed on human blood gave evidence of ". . . a definite quantitative relationship between the weight of blood meal and the number of eggs produced." Woke (1937a) presented data on the same species of mosquito which shows (Table I) that a similar quantitative relationship occurred when the blood of each of seven species of vertebrate hosts including man was ingested. Although the numbers of mosquitoes used are low in most cases, the trend shown is toward increasing numbers of eggs with increasing quantities of blood.

The quantitative relationship must be taken

into account in experimental work in which comparisons are to be made between differently treated groups. The principal anticipated applications are in studies on nutrition and on utilization of the blood meal in ovulation. The information which is provided is needed in setting up experiments by which to more effectively compare the values of different bloods, different components of blood, and different selected nutrients for the development of eggs. Since marked differences are shown in body weights, blood meal weights, and numbers of eggs developed between the strain of *Aedes aegypti* that was used by Roy and the strain that is used in this Laboratory, a determination of values for our strain becomes necessary.

TABLE I

MEAN EGGS DEVELOPED AFTER FIRST BLOOD MEAL FROM 7 DIFFERENT VERTEBRATE SPECIES, RELATED TO QUANTITIES OF BLOOD INGESTED, IN *Aedes aegypti*.[a] AGE: 7 DAYS. BODY WEIGHT: 2.4–2.9 MGS.

Host	Quantities of blood ingested, in milligrams						
	0.5–0.9	1.0–1.4	1.5–1.9	2.0–2.4	2.5–2.9	3.0–3.4	3.5–3.9
Man	.....	.....	64 (2)	78 (10)	94 (22)	100 (2)	100 (2)
Monkey	.....	38 (3)	58 (7)	72 (14)	80 (6)	.....	.....
Rabbit	.....	78 (1)	115 (7)	122 (8)	121 (9)	104 (3)	.....
Guinea pig	38 (1)[b]	80 (4)	105 (13)	112 (11)	130 (3)	.....	.....
Frog	55 (1)	77 (4)	101 (20)	120 (3)	121 (3)	.....	.....
Turtle	42 (2)	88 (1)	92 (13)	107 (8)	111 (5)	.....	.....
Canary	.....	64 (5)	93 (8)	105 (9)	128 (10)	139 (1)	.....

[a]Unpublished data (Woke 1937a).

[b]The number in parenthesis is the number of mosquitoes upon which the mean number of eggs is based.

MATERIALS AND METHODS

The mosquitoes that served as experimental subjects in these experiments were from a long-established laboratory strain of *Aedes aegypti*. This species is suitable for the purpose since normally it develops eggs only after ingesting blood.

All stages of the mosquito were kept in a chamber that was maintained at approximately 28° C. and 70 percent RH, and were continuously exposed to the light from a 4-watt white, fluorescent illuminating tube which gave approximately 4 to 20 foot-candles of illumination around the insects.

The larvae were reared in glass culture dishes on a mixture by weight of one part finely ground commercial chow for laboratory animals[2] and one part brewers' yeast suspended in distilled water at the rate of 2.8 g. per liter. Each dish contained 250 ml. of the water and was approximately 195 mm. in inside diameter at the water surface. The depth of the water was approximately 10 mm. No more than 50 larvae in any single batch were reared in each dish. Daily observations of pH, mortality, and general conditions of the medium and of the larvae indicated no adverse changes in the medium during the development of the larvae. All batches received similar care.

Pupation occurred on the fifth, sixth, and seventh days. The adult females that were chosen as experimental subjects emerged from fifth and sixth day pupae. Approximately 75 females were kept in a community 10 inch cubical cage with the numbers of males that emerged from the same batch, which were approximately equal in number to the females, all during the pre-bloodfeeding period. Five percent sterilized sucrose solution absorbed in cellucotton was provided as food until approximately 20 hours before the time of the blood meal. During the 20-hour period, neither food nor water was provided.

Man was chosen as host because *Aedes aegypti* produces fewer eggs after feeding on human blood than after feeding on similar quantities of the blood of any one of at least five other animals (Woke 1937c). The greatest number of eggs that any individual *Aedes aegypti* can produce on human blood, therefore, is likely to be well under the egg-producing capacity of the species.

The mosquitoes were fed exclusively on the bloods of two young adult human females, one of whom (host A) supplied 500 of the 552 blood meals. Host B supplied the remaining 52 meals. Since no statistically significant difference was found in the egg-production of mosquitoes fed on host A and on host B the data from the two hosts were combined. The effects of only first blood meals were determined. The mosquitoes, as selected, were placed individually in numbered glass feeding cages. For the purpose of feeding, the cages were attached to the skin in a manner which reduced pressure variation on the skin and shaking of the cage (fig. 1).

A required gradation in quantities of blood

[2]"Purina". Ralston Purina Co., St. Louis 2, Missouri.

ingested from near 0.0 mgs. to complete engorgement, was obtained (1) through acceptance of the natural variation in quantities freely ingested, which was satisfactory for quantities of 2.0 mgs. and above, and (2) through intentional interruption of feeding, which was necessary in order to obtain enough mosquitoes ingesting quantities in the lower blood meal weight classes.

Each mosquito was weighed on a 10 mg. capacity Roller-Smith Precision balance within three hours before feeding and within 15 minutes after feeding. It was anesthetized each time with CO_2, then placed on the pan with forceps, weighed, and returned to its feeding cage before recovery. The difference between the pre- and post-feeding weights was accepted as the weight of the blood meal. The mosquito was then released in an individual oviposition cage which consisted of either a lantern chimney set in a half petri dish or a cylinder of celluloid (fig. 2) set over the upper half of a petri dish. A sheet of celluloid was formed into a cylinder and the overlapping edges were joined by the use of acetone. The cylinder was re-inforced by a metal embroidery hoop near each end, the celluloid being held between the two rings that make up each hoop. A net covering was stretched over the inner ring of the upper hoop and secured between the ring and the celluloid wall. The free edges hanging within the cage served as resting places for the insects. The inner lower hoop rested on the edge of the petri dish, while the wall of the celluloid cylinder projected downward around the dish. An adhesive plaster tab was secured to the dish for easy separation of the dish from the cylinder. Each petri dish contained a fitted pad of wet cellucotton covered by a disk of absorbent paper so as to provide the female mosquito a moist surface on which to lay eggs. Sugar solution absorbed in cellucotton was provided in small aluminum cups.

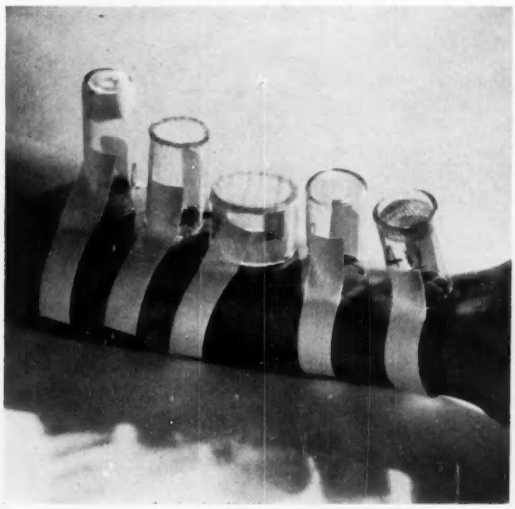

FIG. 1. Feeding cages resting lightly on arm and secured with strips of adhesive plaster. Shown are five types of cages that are useful in feeding.

Undeposited eggs, all of which were full sized and normal in appearance, were removed from the ovaries after two weeks or at death of the insect. They were then counted and included with counts of deposited eggs in the total count of developed eggs. That these unlaid eggs were complete and viable is indicated by results of experiments reported by Woke and Brooks (In preparation). The immediate laying of eggs that probably would have otherwise been deferred (Woke 1955) was induced by the application of mechanical stresses. The deposited eggs appeared normal in every respect. The hatch was normal and comparable to that of eggs laid by the same mosquitoes before the application of the stress. The inculsion of long-retained eggs with normally laid eggs in counts of those fully developed, therefore, seems justified.

FIG. 2. Celluloid oviposition cage.

Hereafter, the age of the mosquito (in days after eclosion) at the time the blood meal was ingested, will be referred to as "age," the weight of the mosquito (in mgs.) before the blood meal was ingested will be referred to as "body weight," and the weight of the blood ingested (in mgs.) will be referred to as "blood meal weight."

INFLUENCE OF HOST-ASSOCIATED FACTORS ON THE NUMBER OF EGGS DEVELOPED

That different host species differentially affect the numbers of eggs that are developed has already been shown (Woke 1937b, c). For the correct interpretation of the results reported in this article it is necessary to consider that certain other host-associated factors might produce differential effects. The available data, therefore, were analyzed to provide the needed information, although they are insufficient to justify universally applicable conclusions.

Variation in effects of different bloods on egg development might arise from variations in the kinds and proportions of the different blood constituents between hosts of the same or different species, sex, age, or physical condition (Albritton 1952, Buxton 1948, DeMeillon and Hardy 1951, Woke 1937c, Yoeli and Mer (1938).

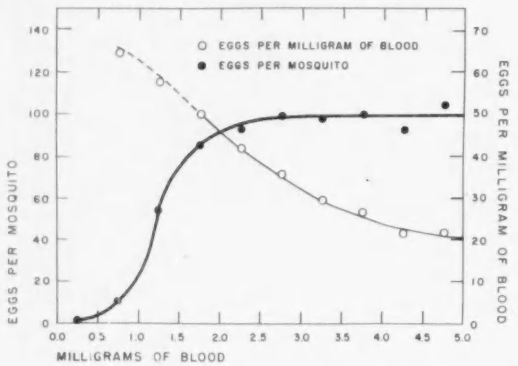

FIG. 3. Numbers of eggs developed by *Aedes aegypti* per mosquito and per milligram of blood ingested, related to quantities of human blood ingested. Age: 7 days. Body weight: 2.5–2.9 mgs. The broken line portion of the curve that represents "Eggs per milligram of blood" is based on the quantities of ingested blood after exclusion of those mosquitoes that laid exceptionally few or no eggs; see footnotes a-c, Table IV. The plotted points represent averages.

Differences in the blood of a single host between times of mosquito feeding. No significant differences were found in the numbers of eggs that were developed by nine groups of 3 to 25 mosquitoes each, fed at widely separated times over a period of 22 months on host A. The mean number of eggs for the total of 73 mosquitoes was 94 and the range was 90 to 98. The nature and extent of variations in the host's blood were not determined since the work was not planned with this objective in mind. The mosquitoes were 7 days old at the time of the blood meal and weighed 2.0–2.9 mgs. They ingested 2.0–2.4 mgs of blood.

Woke (1937c) found no significant differences in the numbers of eggs developed between groups fed on a human host who (1) has fasted for 12 hours overnight, (2) had fasted for 24 hours, and (3) had ingested a full meal within 1½ hours.

Individual differences between hosts. No significant differences are apparent from the egg-production records of mosquitoes which had been fed on two human females. In the first of two series of comparisons, the mosquitoes were 7 days old at the time of the blood meal, weighed 2.0–2.4 mgs., and ingested 2.6–3.3 mgs. of blood per mosquito. Fifteen ingested blood of host A and developed an average of 87 eggs each. Seven ingested blood of host B and developed an average of 88 eggs each.

In the second series the mosquitoes were 28 days old at the time of the blood meal, weighed 2.5–2.9 mgs., and ingested 2.3–4.2 mgs. of blood. Nine ingested blood of host A and produced an average of 57 eggs each. Five ingested blood of host B and developed an average of 55 eggs each.

These results corroborate those of Woke (1937c) for human male blood.

Sex differences between hosts. From unpublished data presented by Woke (1937a) we find that 22 mosquitoes fed on the blood of a young adult human male, host W, and produced an average of 94 eggs. From the data currently reported, we find that 37 mosquitoes fed on the blood of a young adult human female, host A, and produced an average of 96 eggs.

The mosquitoes were 7 days old at the time of the blood meal and weighted 2.4–2.9 mgs. They ingested between 2.5 and 2.9 mgs. of blood each.

Since no significant difference in effects of the two bloods was apparent, we are enabled to make direct comparison of the data obtained in the two studies.

INFLUENCE OF ARTHROPOD-ASSOCIATED FACTORS ON THE NUMBERS OF EGGS DEVELOPED

Individual differences. The numbers of eggs that were developed by the different mosquitoes ranged from 0 to 136. That differences between individuals account for much of this wide variation will be apparent from an inspection of the tables which follow.

Age. The relation of the age of the mosquito to the numbers of eggs developed is summarized in Table II. The actual differences in the average numbers of eggs developed by mosquitoes aged 5, 6, 7, and 14 days do not appear significant, but these younger mosquitoes clearly developed significantly more eggs than did those aged 28 days. In selecting mosquitoes for use in comparative studies involving egg-production, these results indicate that little importance need be attached to age differences between 5 and 14 days. Since no problem is involved in the matter of selecting ages, one might choose, with advantages, any age within this range. If one chooses seven-day old mosquitoes, the period from eclosion to age seven days gives ample

time for the fullest development of the ovaries and ovarian follicles. The high level of egg production found 7 days later gives assurance that the age seven days is well under that at which important reduction in numbers of eggs produced, due to age, is likely to occur.

Body Weight. The frequency distribution of body weights of female mosquitoes produced in the laboratory cultures was as follows: Body weight class 1.5–1.9 mgs. (actually only 1.8–1.9 mgs.), 6 mosquitoes; 2.0–2.4, 125 (30 percent); 2.5–2.9, 171 (40 percent); 3.0–3.4, 111 (26 percent); and 3.5–3.9, 12.

The relation of the body weight to the numbers of eggs developed is summarized in Table III. While the increases in mean numbers of eggs developed suggest a gradient, the differences from class to class are small and a statistically significant trend could not be established owing to the extremely large variation in the numbers of eggs developed by the individual mosquitoes in each group.

TABLE II

RELATION OF AGE OF THE MOSQUITO AT THE TIME OF THE BLOOD MEAL TO THE NUMBERS OF EGGS DEVELOPED. *Aedes aegypti*. BLOOD MEAL WEIGHT: 2.0–2.9 MGS. BODY WEIGHT: 2.5–2.9 MGS.

Age (days)	Number of mosquitoes	Eggs per mosquito		
		Range	Mean	S.D.
5	26	54–112	89	17
6	19	39–118	85	21
7	71	66–125	95	14
14	13	64–112	86	15
28	18	28– 79	56	12

The lack of demonstrated statistical differences may remove need for exacting care in the selection of mosquitoes for experimental work, especially work in which expected effects are different to a highly significant degree. However, restriction to body weight class 2.0–2.9 mgs. (the body weights of approximately 69 percent of the reared female mosquitoes fell within this class) or to an even narrower class 2.5–2.9 mgs. (the body weights of approximately 40 percent fell within this class) offers no special problem. Adequate numbers are assured and any doubt in regard to the possible effects of body weight is reduced.

The relation between body weight and blood meal weight was explored statistically but found negligible.

Roy (1936) concluded that the weight of the mosquito is not the determining factor in the numerical productivity of eggs, but that the amount of blood in its stomach probably exercises a greater influence. Shannon and Hadjinicalao (1941) reported a definite positive correlation between the size of individual anophelines and the number of eggs produced.

Blood meal weight. The quantities of blood that the 552 mosquitoes of all ages and body weights ingested, ranged from 0.1 to 4.8 mgs. Of the 165 that took 1.9 mgs. or less, a large number had been intentionally interrupted in their feeding. The frequency distribution of the quantities taken by those mosquitoes that fed to repletion is of interest. Only one is known to have withdrawn voluntarily after having ingested less than 0.5 mg. of blood; this one took 0.3 mg. Not more than 5 voluntarily limited themselves to quantities between 0.5 and 0.9 mg. A much larger number, 98 or fewer, freely ingested quantities between 1.0 and 1.9 mgs. Three hundred eighty-seven mosquitoes freely ingested 2.0 mgs. or more each. The frequency distribution is: blood meal weight class 2.0–2.4 mgs., 121 mosquitoes (31 percent); 2.5–2.9, 131 (34 percent); 3.0–3.4, 70; 3.5–3.9, 50; 4.0–4.4, 12; and 4.5–4.9, 3. The distribution is essentially the same in each age and body weight group. The two blood meal weight classes, 2.0–2.4 and 2.5–2.9 mgs., together account for 252 or 65 percent of the mosquitoes that fed freely.

TABLE III

RELATION OF THE BODY WEIGHT OF THE MOSQUITO, *Aedes aegypti*, TO THE NUMBERS OF EGGS DEVELOPED. BLOOD MEAL WEIGHT: 2.0–2.9 MGS.

Age (days)	Body weight mgs.	No. of mosquitoes	Eggs per mosquito		
			Range	Mean	S.D.
5–6	2.0–2.4	27	46–107	82	17
	2.5–2.9	45	39–118	87	19
	3.0–3.4	17	40–129	94	23
7	1.5–1.9	6	59–109	84	17
	2.0–2.4	23	63–108	89	13
	2.5–2.9	71	66–125	95	14
	3.0–3.4	22	42–111	88	17

The quantities of blood that are ingested may have significance not only for egg production, but also for rates of infection with the causative agents of human and animal diseases. Differences in quantities may in part account for differences in infection rates.

The relations between the numbers of eggs developed and the quantities of human blood ingested by 7-day old mosquitoes in the 2.5–2.9 mg. body weight class is given in Table IV and fig. 3. That for 5- and 6-day old mosquitoes in the 2.5–2.9 mg. body weight class, and 6- and 7-day old mosquitoes in the 3.0–3.4 mg. body weight class, though based on small numbers, showed similar trends.

The failure to develop eggs on the part of 55 of the 82 mosquitoes that ingested 1.4 mgs. or less, and the development of exceptionally few

eggs (between 1 and 11) on the part of 6 other mosquitoes, (see Table IV and footnotes a-c) is clearly related to the small quantities of blood ingested; these mosquitoes were regarded as normal.

The numbers of eggs developed by each of the remaining 21 mosquitoes that took 1.4 mgs. or less are comparable, in terms of eggs per mg. of ingested blood, to the numbers that were usually developed by mosquitoes that took greater quantities. The rapidly increasing proportion of mosquitoes that developed "usual" numbers of eggs, is clearly related to the increasing sizes of the blood meals.

The quantities of blood ingested by the 13 mosquitoes that developed eggs after ingesting less than 1.0 mg. of blood, and the numbers of eggs that were developed, are as follows: 0.1 mg., 2 eggs; 0.3 mg. 4; 0.4 mg., 23; 0.5 mg., 1; 0.6 mg., 3 and 49; 0.8 mg., 11, 44, and 62; 0.9 mg., 2, 47, 52, and 55. In the experiments of Roy, 0.82 mg. was the lowest amount of blood upon which eggs were produced.

In order to detect small differences in egg production between differently treated groups, mosquitoes must be chosen in such a way as to reduce the within-group variation; comparisons must be made only between groups that have ingested similar quantities of blood with similar within-group distributions.

TABLE IV

NUMBERS OF EGGS DEVELOPED BY *Aedes aegypti* RELATED TO QUANTITIES OF HUMAN BLOOD INGESTED. AGE: 7 DAYS. BODY WEIGHT: 2.5–2.9 MGS.

Quantities of blood ingested (mgs.)	No. of mosquitoes	Eggs per mosquito			Mean eggs per milligram of blood
		Range	Mean	S.D.	
0.0–0.4	31[a]	0– 23	1	..	..
0.5–0.9	32[b]	0– 62	10	..	..
1.0–1.4	19[c]	0– 91	54	..	..
1.5–1.9	28	43–113	85	16	50
2.0–2.4	44	66–119	93	14	42
2.5–2.9	27	72–125	99	14	36
3.0–3.4	22	60–127	98	16	30
3.5–3.9	14[d]	53–132	100	23	27
4.0–4.4	3	80–105	93	12	22
4.5–4.9	1	.....	105	..	22

[a]Of the 31 mosquitoes 28, (90 percent) did not develop eggs; 1 developed 2, and 1 developed 4. One mosquito developed 23 eggs (49 per mg. of ingested blood).

[b]Of the 32 mosquitoes 22, (69 percent) did not develop eggs; 4 developed a total of 17. Six mosquitoes developed a mean of 51 eggs (range 44–62, S.D. 6.4) (64 per mg. of ingested blood).

[c]Of the 19 mosquitoes, 5 (26 percent) did not develop eggs. 14 developed a mean of 73 eggs (range 47–91; S.D. 14). (58 per mg. of ingested blood).

[d]One additional mosquito did not develop eggs. It is regarded as an anomaly, being the only one among 139 in the group that ingested 1.5 mgs. or more, that did not develop eggs.

In these experiments, using human blood, the mean numbers of eggs that were developed by mosquitoes after ingesting 2.5–2.9 mg. of blood was not exceeded significantly by mosquitoes which ingested larger quantities. The additional increments at most added a very few eggs to the maximum production in each range. Thus we see that the mosquito can and often does ingest more human blood than she uses in developing eggs. Whether the "excess" blood is utilized in some manner or is merely excreted is yet to be determined. The results obtained by Roy show a tendency toward a decreasing rate of production on larger quantities of ingested blood, but no definite plateau such as that shown here.

The most suitable meal-weight range to use in comparative tests may be chosen on the basis of a predetermined estimate of the egg production to be expected from each nutrient and judgement by comparison with the known egg production on human blood. The meal-weight range of choice may be that range which gives near capacity egg production on the nutrient on which highest egg production is expected. For human blood this is 2.5–2.9 mgs. The choice of mosquitoes that have taken 2.5–2.9 mgs. of blood assures a large number of mosquitoes of desirably high egg producing potential. Since over half of the freely fed mosquitoes ingested quantities of 2.0–2.9 mgs., this range assures even greater numbers. The increase in the standard deviation would be too slight to seriously effect the sensitivity of the test.

Recently Lang and Wallis (1956) reported that the inclusion of sucrose in test nutrients stimulates engorgement. Since by this means greater numbers of mosquitoes fully engorge, comparisons can be made on larger quantities of nutrient.

SUMMARY AND CONCLUSIONS

This article reports on relations between the numbers of eggs that are developed by *Aedes aegypti* (Linnaeus) and the quantities of human blood that are ingested as a basis for planned studies on arthropod-vertebrate host interrelations.

Five hundred fifty-two mosquitoes took first and only blood meals at ages either 5, 6, 7, 14, or 28 days. The body weights ranged between 1.5 and 3.4 mgs. The quantities of blood ingested ranged from 0.1–4.8 mgs. The numbers of eggs that were developed by the different individuals ranged from 0 to 136.

The chief conclusions are: No significant differences in numbers of eggs developed were found that could be attributed to differences in the blood of a single host between times of mosquito feeding, to individual differences between two female hosts, or to sex differences between two hosts. For mosquitoes in these experiments given the blood meal on the seventh

day after emergence, the mean numbers of eggs that were developed ranged from 87 to 98, with wide individual variation.

Mosquitoes that took the blood meal at age 28 days developed significantly fewer eggs (mean 56) than those that took the blood meal at ages 5, 6, 7, and 14 days (means 89, 85, 95, and 86 respectively). The latter age groups are not regarded as significantly different from each other.

The actual mean numbers of developed eggs increased as the mosquito body weight increased. Although the increase suggests a normal gradient, a statistically significant trend was not established.

A very marked upward trend in egg production (from a mean of 1 egg to a mean of 99 eggs) is correlated with successively larger quantities of ingested blood (each with a 0.5 mg. range) up to around 2.5–2.9 mgs., where the curve levels off. Beyond this point the only trend seen is a small but constant increase in the maximum number of eggs developed, to 132 eggs after 3.5–3.9 mgs. of blood.

In experimental studies involving the egg/nutrient quantitative relation, an age of 7 days at the time of the meal, with body weights restricted to the range 2.0–2.9 mgs., or, preferably, 2.5–2.9 mgs., with the meal-weight range that range which gives near capacity egg production on the nutrient on which highest egg production is expected, and with distribution of weights within compared groups equalized, should be suitable for detecting significant differences between compared groups.

A basis is prepared for experimentation into requirements for ovulation and egg production, origin and evolution of the blood-feeding habit, and behavior in relation to hosts.

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PRELIMINARY STUDIES OF SUGARS AND AMINO ACIDS IN THE HONEYDEWS OF FIVE SPECIES OF COCCIDS FEEDING ON CITRUS IN CALIFORNIA[1]

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Deposits of crystalline or syrupy honeydews (or mannas) on plants have attracted attention since ancient times. (See Braun, 1938, for a review of early literature.) Essig (1942) points out that the "manna of the Wilderness" of Biblical accounts was honeydew produced by the coccid *Trabutina mannipara* (Ehr.), feeding on Tamarix trees.

It is customary to refer to such deposits as mannas if they are plant secretions and as honeydews if they are produced by insects. Hudson (1946) has indicated, however, that the origins of some so-called "mannas," notably the crystalline deposits on camelthorn in Turkistan and on Douglas fir in Canada, have not been fully clarified, and further studies may yet show that they are insect-produced and should be properly classified as true honeydews.

Apiarists have long known that in the absence of floral nectars honeydews are collected by bees. Honeys derived from certain honeydews, including those produced by the aphids *Cinara pectinatae* (Nördl.) (=*Lachnus pichtae* Mordv.) on fir and *C. piceae* (Panz.) (=*Lachnus grossus* Kalt.), on spruce in central Europe have considerable economic value (Braun, 1938; Geinitz, 1939; Gontarski, 1940); others, notably the so-

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called "melezitose honey," derived from deposits associated with the aphid *Schizolachnus* (=*Lachnus*) *pineti* (Fabr.) and the coccid *Toumeyella parvicorne* Ckll. on pine in the eastern United States, are of very poor quality (Hudson and Sherwood, 1920; Hudson, 1946).

Honeydew production is not well understood. Insects excreting this type of material feed by means of elongated, slender mouthtubes from internally located plant tissues. A part of the alimentary system of these insects consists of a specialized structure known as the filter chamber, which varies considerably in shape and complexity in different species (Pesson, 1934, 1935, 1936; Michel, 1942). Morphological studies suggest that this structure has an absorption function, as first proposed by Berlese (1893). This, however, has not been demonstrated, nor is it known what connection, if any, the filter chamber has in the production of honeydew.

A striking feature of honeydew production is the copious amounts produced in relation to the small size of the insects involved. Michel (1942), in studies on *Lachnus roboris* (L.) on oak, measured an average of 5.0 and as much as 8.6 milligrams per aphid during a 24-hour period. This maximum amount is considerably more than the weight of the insect producing it. Day and Irzykiewicz (1953) calculated from studies with radio-phosphorus that *Myzus persicae* (Sulz.) may ingest 35 percent of its weight in one hour. Honeydews are also remarkable in that water-soluble carbohydrates, mainly sugars, may exceed 80 percent of the total weight of the freshly excreted material. The remaining portion is principally water, nitrogen-containing compounds, and traces of other materials.

Attempts have been made to explain the excretion of such large amounts of highly nutritious material. Busgen (1891) suggested that since plant sap has a low nitrogen content relative to the carbohydrates, large amounts must be ingested by the insects to satisfy their nitrogen requirements, with unassimilated material being excreted as honeydew. Berlese (1893) thought that the filter chamber was involved in the absorption of nitrogen. This explanation for such large amounts of excretory material is not entirely satisfactory, for certain insects at least, since appreciable quantities of many of the essential amino acids have been detected in the honeydews of the mealybug *Pseudococcus brevipes* (Ckll.) on pineapple (Gray, 1952), and of the aphids *Myzus circumflexus* (Buck.) on potato (Maltais and Auclair, 1952) and *Tuberolachnus salignus* (Gmel.) on willow (Mittler, 1953).

Recently, another interesting theory was advanced by Kennedy and Mittler (1953) to explain the production of honeydew by *Tuberolachnus salignus* on willow. These investigators found that if the mouthtube of an actively feeding aphid was carefully severed, a considerable amount of fluid continued to ooze from the cut end of the tube remaining in the plant, as a result of pressure within the plant tissues. It is therefore suggested that during the feeding process, plant pressure forces large amounts of sap, in the form of honeydew, through the insect's alimentary system. Whether this explanation is satisfactory for other species of insects is not known. In an earlier study Leonardt (1940) reported an apparent relationship between osmotic pressure variations in fir trees and the production rate of honeydew by *Cinara pectinatae*. Michel (1942) could not verify this, nor were Gray and Fraenkel (1954) successful in obtaining a sufficient amount of fluid for chromatographic studies from severed mouthtubes of the mealybug *Planococcus citri* (Risso) on potato sprouts.

The composition of honeydews has been the subject of many studies. Early investigators, using standard analytical methods, made determinations of water, total nitrogen, and certain readily separated carohydrates, including glucose, fructose, sucrose, melezitose, and dextrin. Among these studies were analyses of honeydews from *Chaetaphorella aceris* (L.) on maple, *Cinara laricis* (Ckll.) (=*Lachnus muravensis* Arnh.) on larch, *Trabutina mannipara* (Ehr.) on tamarix, *Cinara pectinatae* (Nördl.) on fir, *Myzocallis tilliae* (L.) on linden, *Pemphigus spirothacae* Pass. on various plants, *Cinara piceae* (Panz.) on spruce, *Physokermes piceae* (Schr.) (=*Lecanium hemicryphum*) on spruce, and *Lachnus roboris* L. on oak (Arnhart, 1926; Weber, 1930; Toth, 1933; Gontarski, 1940; Michel, 1942). Additional investigations have been conducted on "mannas," not associated with insects, from larch, camelthorn, and Douglas fir, and on honeys derived from honeydews associated with *Cinara laricis* on larch (Arnhart, 1926) and *Toumeyella parvicorne* and *Schizolachnus pineti* on pine (Hudson, 1942).

Later investigators working with chromatographic techniques have separated and identified various amino acids, carbohydrates, and other components not readily separated with previously used chemical methods. Gray (1952), on the basis of position of ninhydrin spots on two-dimensional chromatograms, reported 19 free amino acids in the honeydew of *Pseudococcus brevipes* on pineapple. These included cystine, aspartic acid, glutamic acid, serine, glycine, asparagine, glutamine, alanine, tyrosine, histidine, tryptophane, phenylalanine, proline, and three unidentified spots.

Carbohydrate components included glucose, fructose, sucrose, glucose-l-phosphate, and possibly maltose. Malic acid, citric acid, and salts of the latter were also reported. Maltais and Auclair (1952) reported 22 amino acids in the honeydew of *Myzus circumflexus* on potato, including *alpha* and *beta*-alanine, *alpha*-amino-*n*-butyric acid, *gamma*-amino butyric acid, argin-

ine, asparagine, aspartic acid, cysteic acid, glutamic acid, glutamine, glycine, histidine, leucine and/or isoleucine, lysine, methionine sulfoxide, phenylalanine, proline, serine, theoreonine, tryptophan, tyrosine, and valine. Quantitative studies showed this honeydew to contain (on dry-weight basis) 13.2 percent amino acids and 35.7 percent invert sugar.

Hackman and Trikojus (1952) reported the presence of ribitol in honeydews produced by *Ceroplastes destructor* Newst. on lemon and *Bursaria*; by *Ceroplastes rubens* Mask. on citrus, mango, gordonia tree, and *Eugenia;* by *Saissetia oleae* (Bern.) on orange; and by *Eriococcus eucalypti* Marsh on *Bursaria*. This compound was not found in deposits produced by *Eriococcus coriaceus* Marsh on *Eucalyptus*. Other reports of sugar alcohols include dulcitol in honeydew of *Aphis euonymi* Fab. on *Euonymus* (Molliard, 1931) and mannitol in camelthorn "manna" (Hudson and Sherwood, 1918). It is generally believed that sugar alcohols in plant sap are passed unchanged through the insect's alimentary system. Gray and Fraenkel (1953, 1954) found, in the honeydew of *Planococcus citri* on potato sprouts, fructose, glucose, sucrose, glucose-l-phosphate, and a trisaccharide referred to as fructomaltose. Since this last-named sugar was not detected in potato sprout sap, it is concluded by these authors that it is an intermediate product formed in the process of hydrolysis of sucrose by invertase produced by the insect. Fructomaltose was also found in the honeydews of *Aphis spiraecola* Patch and *Pulvinaria vitis* (L.), as well as in honey, and apparently is identical with the trisaccharide α -maltosyl β -D-fructofuranoside, synthesized by White and Maher (1953).

Likewise, Michel (1942), in studies of *Lachnus roboris* on oak, concluded that melezitose was a synthetic product of the insect, since this sugar was not detected in the sap of the host. Mittler (1953) found in the honeydew of *Tuberolachnus salignus* on willow the following amino acids: aspartic acid, glutamic acid, serine, threonine, alanine, asparagine, glutamine, valine, leucine and/or isoleucine, phenylalanine, and probably *gamma*-amino butyric acid. All amino acids in the honeydew were found in willow sap, indicating that the insects do not differentially absorb these compounds from the host plant.

The foregoing studies indicate that honeydews are very complex in composition, containing not only unchanged plant sap materials, but synthetic components as well. There is also evidence that the composition varies with the insect species and host plant. Another manifestation of dissimilarity in the composition of honeydews is their variability in attractiveness to other insects (Schmutterer, 1953). Bees, for example, are readily attracted to honeydew deposits on spruce, fir, pine, linden, beech, and elm, but not to those on *Sambucus*, *Philadelphus*, and plum.

A number of the common insect pests of citrus are honeydew producers. Among those occurring in California are cottony-cushion scale, *Icerya purchasi* Mask.; soft scale, *Coccus hesperidum* L.; citricola scale, *C. pseudomagnoliarum* (Kuw.); black scale, *Saissetia oleae* (Bern.); citrus mealybug, *Planococcus citri* (Risso); grape mealybug, *Pseudococcus maritimus* (Ehrh.); long-tailed mealybug, *P. adonidum* (L.); citrophilus mealybug, *P. gahani* Green; and several species of aphids, principally *Aphis spiraecola* Patch, *A. medicaginis* Koch, *A. gossypii* Glov., *Myzus persicae* (Sulz.), and *Toxoptera aurantii* (Fonsc.).

The honeydew of *Icerya purchasi* invariably crystallizes within a few hours after it is excreted, while those produced by the other species normally occur as sticky syrups, only rarely forming crystals. Observations in the field indicate that the syrupy deposits are not all alike in composition since the Argentine ant, *Iridomyrmex humilis* Mayr., readily seeks out colonies of *Coccus hesperidum*, apparently preferring the honeydew of this species to that of other coccids. Conversely, deposits from *C. pseudomagnoliarum* apparently have little or no attractiveness to this species of ant.

Recently, investigations were undertaken in our laboratory to determine whether qualitative differences in composition occurred among honeydews produced by certain coccids commonly found on citrus. It is thought that information of this type would not only be of value in the study of insect attractants, but would also contribute to a better understanding of nutritional requirements of some of the most important insect pests of citrus. It is also conceivable that information on honeydew components could ultimately be useful in studies on the mode of action of insecticides, particularly those having systemic characteristics. This report is a summary of preliminary studies of the sugar and amino acid components.

MATERIALS AND METHODS

Included in this investigation were the honeydews of *Icerya purchasi*, *Coccus hesperidum*, *Planococcus citri*, *Coccus pseudomagnoliarum*, and *Saissetia oleae*. For most of the work, rooted lemon cuttings and grapefruit seedlings grown in the greenhouse served as host plants for the first three species named above. For purposes of comparison a few samples of honeydew were collected from citrus trees in commercial groves, and, in the case of *P. citri*, from orange fruit kept in the greenhouse. All samples of honeydew for the last two species named above were collected from the field on commercially grown navel orange trees. Collections were made from accumulations on leaves and twigs or on glass slides placed beneath the insects, care being taken to exclude material showing sooty-mold fungus growth. The samples were diluted with

distilled water to final solutions containing from 5 to 25 percent honeydew, filtered to remove exuviae and other debris, and stored under toluene in a refrigerator. As soon as any deviation in composition from the freshly collected material was detected on chromatograms, the samples were discarded.

The water content of several samples of honeydews from *Icerya purchasi*, *Coccus hesperidum*, and *Planococcus citri*, collected from both field and greenhouse, was determined by heating to constant weight in an oven at 70° C. Under these conditions the dried materials turned slightly brown, thus indicating that some caramelization occurred.

Investigations of the sugars and amino acids were conducted by means of paper-partition chromatography, a method well adapted for studies with limited amounts of material. For most routine work with one-dimensional, ascending chromatograms the various honeydews and standard solutions were spotted 1 inch apart on the starting line, ¾ inch from the edge of a sheet of Whatman No. 1 filter paper measuring 8½ inches square. After the spots had been air-dried, the paper was formed into a cylinder by stapling in juxtaposition the two edges perpendicular to the starting line. These cylinders were then placed in an upright position in standard petri dishes containing appropriate developing solvent, and were allowed to develop at 15° C. in tightly capped bell jars approximately 10 inches in diameter and 18 inches high. Two-dimensional chromatograms were run in a similar manner, except that the single spot was placed in the corner of the paper 1 inch from each edge. Occasionally, when more complete separation of the spots was desired, the descending technique was used on 18-inch-square papers.

The developing solvent for separating sugars consisted of the upper phase of a mixture of 2 parts pyridine, 3 parts *n*-butanol, and 1.5 parts water (by volume), made according to the directions of Chargoff *et al.* (1948). One to 5 microliters of solutions containing from 5 to 25 percent honeydew were spotted on the starting line along with similar amounts of known solutions containing various mixtures of fructose, glucose, sucrose, maltose, and melezitose. The papers were double-developed (Jeanes *et al.*, 1951) for approximately 12 hours for each development, with no attempt being made to stop the solvent front before it reached the edge. When air-dried for several hours, sprayed with aniline hydrogen phthalate reagent (Partridge, 1949), and heated for 3 minutes at 105° C., the aldoses, glucose and maltose, appeared as brown spots. The papers were then over-sprayed with a reagent modified from that of Shriner and Fuson (1944) and made up by stirring into 75 ml. of ethyl alcohol a solution containing 1 gram 2, 4-dinitrophenylhydrazine in 7.5 ml. concentrated sulfuric acid, and then further diluting with 250 ml. of ethyl alcohol. After spraying, the papers were air-dried and heated for 1 to 2 minutes at 105° C., the keto-function sugars fructose, sucrose, and melezitose then appearing as bright-yellow spots. The known sugars had the following approximate R_s values: fructose 1.48, glucose 1.24, maltose 0.79, melezitose 0.56.

Amino acids were separated on two-dimensional chromatograms, the first solvent being the pyridine-*n*-butanol-water mixture used in separating sugars, and the second solvent being the upper phase of a mixture of 1 part acetic acid, 4 parts *n*-butanol, and 5 parts water (by volume). The papers were double-developed in each direction, air-dried for several hours, and sprayed with a solution of 0.5 percent ninhydrin in *n*-butanol. The spots were marked after the papers were air-dried and heated at 105° C. for 3 minutes.

RESULTS

The amount of water removed by heating at 70° C. from the honeydews of *Icerya purchasi*, *Coccus hesperidum*, and *Planococcus citri* varied somewhat, depending on the age of the samples and environmental humidity at time of collection but never exceeded 15 percent of the weight of the freshly excreted materials. Average amounts removed from several samples produced by these three species under both greenhouse and field conditions amounted to 10.39, 9.16, and 11.30 percent, respectively.

Sugar spots from typical samples of honeydews on citrus are shown on the reconstructed chromatogram (fig. 1). Four sugars occurred in the honeydew of *Icerya purchasi* and six in those from the other four species. This pattern of sugar components was remarkably constant, regardless of whether the samples were collected in the field or in the greenhouse, or whether the host plant was lemon, grapefruit, or orange; variations in samples from insects feeding on leaves or twigs and branches (or fruit in the case of *Planococcus citri*) also appeared to be insignificant. Sugars Nos. 1, 2, and 3, corresponding to fructose, glucose, and sucrose, respectively, occurred in samples from all five species studied. Of special interest is the glucose spot, which is relatively large in the honeydew of *P. citri* and small in the other species, indicating quantitative differences. Judging from the size of the glucose spots from a large number of samples of honeydews produced under both field and greenhouse conditions by *I. purchasi* and *C. hesperidum*, it appears that, for these two species at least, this sugar is normally present in relatively small amounts. Actually, in some samples a glucose spot failed to appear on the chromatograms. The significance of the relatively small amounts of this sugar is not understood at the present time.

There was also present in all honeydew samples

a sugar (No. 4) having an R_s corresponding to melezitose. In the case of *Icerya purchasi* this sugar was separated on a cellulose column (Gross and Albon, 1953), and several milligrams were collected in the crystalline state. It was found to be indistinguishable from melezitose in that it had an identical specific rotation

$$(\alpha)_D^{20} = +88.2, \text{ m.p. } 153\text{–}4^\circ \text{ C.},$$

was nonreducing, crystallized very readily, and was not hydrolyzed by yeast invertase. It is the principal sugar in, and apparently responsible for the crystalline nature of, the honeydew of *I. purchasi*. This interesting trisaccharide is known

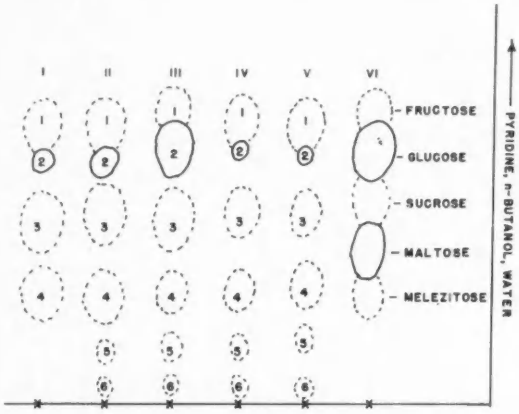

FIG. 1. Reconstructed chromatogram of sugars in typical honeydews of species of coccids feeding on citrus:
I. Cottony-cushion scale, *Icerya purchasi* Mask., on grapefruit leaf.
II. Soft scale, *Coccus hesperidum* L., on grapefruit leaf.
III. Citrus mealybug, *Planococcus citri* (Risso), on Valencia orange fruit.
IV. Citricola scale, *Coccus pseudomagnoliarum* (Kuw.), on navel orange twig.
V. Black scale, *Saissetia oleae* (Bern.), on navel orange leaf.
VI. Standard solution of known sugars.
Paper sprayed first with aniline hydrogen phthalate reagent to show aldo-function sugars (solid lines) and then oversprayed with 2, 4-dinitrophenylhydrazine reagent to show keto-function sugars (broken lines). One microliter of a 25 percent solution of each honeydew was spotted on the starting line.

to have a widespread occurrence in honeydews and "mannas," having been previously recorded from many deposits, including those on larch, fir, pine, poplar, linden, bamboo, oak, camelthorn, and Douglas fir (see Hudson, 1946).

The principal sugar (No. 4) in the honeydew of *Coccus hesperidum* has been shown by Wolf and Ewart (1955a) to be another trisaccharide, gluco-sucrose. This is apparently identical with the fructomaltose found in the honeydews of *Planococcus citri* and other insects by Gray and Fraenkel (1953). The work of Wolf and Ewart (1955a) also shows the presence of at least three keto-function sugars (only two of which appeared on the chromatograms in this study) having lower R_s values than gluco-sucrose. These appear to be a polymer-homologous series of oligosaccharides resulting from transglucosylation on sucrose.

The presence of relatively large amounts of melezitose in the honeydew of *Icerya purchasi*, and of gluco-sucrose along with heavier oligosaccharides in that of *Coccus hesperidum*, apparently results from syntheses by the insects, since these sugars could not be detected in extracts of the host plants. This agrees with the conclusions of Michel (1942) with respect to melezitose from *Lachnus roboris* on oak, and of Gray and Fraenkel (1954) with respect to gluco-sucrose from *Planococcus citri* on potato sprouts. Supporting evidence for the synthesis of oligosaccharides in insects comes from the recent isolation of sugar-free, enzyme-containing fractions from honeydews of *I. purchasi* and *C. hesperidum* capable of producing melezitose and gluco-sucrose, respectively, from sucrose (Wolf and Ewart, 1955b).

In deposits from *Planococcus citri*, *Coccus pseudomagnoliarum*, and *Saissetia oleae*, sugar No. 4 as well as the heavier oligosaccharides are, qualitatively at least, chromatographically identical with the sugars found in the honeydew of *C. hesperidum*. A report on detailed studies of these honeydews is in preparation.

A large number of samples of honeydews from the five species of coccids included in this study were examined for amino acids and found to be chromatographically identical, at least qualitatively, regardless of species of insect, variety of host plant, part of plant the insect was feeding upon, or of host location, in the field or in the greenhouse. On the reconstructed chromatogram (fig. 2) are shown 10 ninhydrin spots from the honeydew of *Icerya purchasi* on a grapefruit seedling grown in the greenhouse. These spots, identical for all five species, correspond to arginine, aspartic acid, glutamic acid, asparagine, glutamine, serine, threonine, alanine, proline, and valine. An interesting question is whether the amino acids in the honeydew are identical with those in the sap of the host plant, as was found to be the case with *Tuberolachnus salignus* on willow by Mittler (1953). The answer to this question awaits the outcome of studies not yet completed. The work of Underwood and Rockland (1953) has a bearing on this subject, as they found in grapefruit juice the same amino acids listed above, with the exception that *gamma* amino-butyric acid was present, but not threonine or valine.

Of special interest is the similarity in the sugar and amino acid components of the honeydews of *Coccus hesperidum* and *C. pseudomagnoliarum* in

view of the frequent observation that the Argentine ant is readily attracted to the former but not to the latter. On the basis of this study it appears that this difference in attractiveness is not due to qualitative differences in sugars or amino acids.

SUMMARY

Chromatographic studies were conducted on the sugar and amino acid components of the honeydews of five species of coccids commonly found on citrus in California. The principal sugar in the honeydew of the cottony-cushion scale, *Icerya purchasi* Mask., was found to be melezitose, a trisaccharide formerly reported from a wide variety of honeydews and "mannas."

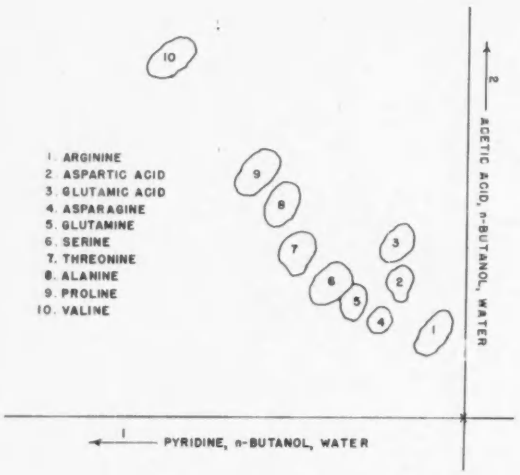

FIG. 2. Reconstructed chromatogram of amino acids in honeydew of cottony-cushion scale, *Icerya purchasi* Mask., feeding on grapefruit leaf. Three microliters (1-microliter increments) of a 25 percent solution of honeydew were spotted on starting line.

Other sugars present included fructose, glucose, and sucrose. Deposits from soft scale, *Coccus hesperidum* L.; citricola scale, *C. pseudomagnoliarum* (Kuw.); citrus mealybug, *Planococcus citri* (Risso); and black scale, *Saissetia oleae* (Bern.), were chromatographically identical with respect to sugar components. The principal sugar was another trisacchraide, gluco-sucrose. Others present included fructose, glucose, sucrose, and at least two oligosaccharides having lower R_s values than gluco-sucrose. The presence of trisaccharides and heavier oligosaccharides appears to be the result of syntheses by the insects, as these sugars were not detected in the host-plant extracts. Ten amino acids, apparently identical for the honeydews of all five species, were chromatographically identified as arginine, aspartic acid, glutamic acid, asparagine, glutamine, serine, threonine, alanine, proline, and valine. Qualitative differences in the sugar and amino acid components apparently are not responsible for the difference in attractiveness the honeydews of *C. hesperidum* and *C. pseudomagnoliarum* have for the Argentine ant, *Iridomyrmex humilis* Mayr.

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LIFE HISTORY STUDIES OF PEPSIS AND HEMIPEPSIS WASPS IN CALIFORNIA (HYMENOPTERA, POMPILIDAE)[1]

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PREFACE

Wasps of the genera *Pepsis* and *Hemipepsis* are our largest hymenoptera. A large *Pepsis* is about an inch and a half long; the tarantulas, their common prey and provision for the young of these wasps, are our largest spiders. Encounters between "tarantula hawks", as these big wasps are frequently called, and tarantulas have often been observed and occasionally recorded in literature. But there has not been much information on what species of wasp attacks what species of tarantula, nor on some of the aboveground operations subsequent to these encounters. This paper supplies some of these data, and partly fills in the almost complete void hitherto existing on the early stages of these pepsine wasps, as well as the operations of the mother wasp in the dark underground where she prepares the way for her offspring that she will never see.

My observations include the summers of 1952, 1953 and 1954, a part of the summers of 1937 and 1951, as well as numerous off-season studies in the laboratory. The living material involved numbers more than two dozen wasps and many more tarantulas. Of these wasps, 15 *Pepsis* and 4 *Hemipepsis* parasitized from one to ten tarantulas apiece, and 30 cocoons, representing the 6 species of wasps used, were produced. Selected activities of most of these wasps are considered in this paper.

Studying wasps and tarantulas under artificial conditions should be supplemented by extensive field observations, where a great deal remains to be done. At this point I wish to thank Mr. Harry A. Hill, of San Diego, for so generously submitting his field observations on a *Pepsis mildei* wasp, which are recorded here in condensed form under the proper heading. I wish also to thank Dr. Edward S. Ross, Curator of Entomology at the California Academy of Sciences, San Francisco, for some of the photographs used in this paper.

Nearly all the material for this paper has been collected by myself, but I owe thanks to those kind people who have contributed a tarantula or two.

The Life History of a Pepsis Wasp

In midsummer the great hairy *Aphonopelma* tarantula paralyzed by the sting of the *Pepsis* wasp lies in a cell in the stony ground. However, for the present study of the development of *Pepsis*, based mainly on *Pepsis thisbe* Lucas, I use live laboratory material. Now, very carefully, I dig a recently parasitized tarantula out of its cell in the large jar. Then I lower it gently onto a shallow bed of nearly dry soil, already pressed down in a suitable container, such as a plastic refrigerator dish that can be covered. Here the delicate egg and the forthcoming grub or larva may be viewed under magnification.

THE EGG

The gently curved creamy white egg scarcely five millimeters long, glued by its slightly thicker end to the tarantula's abdomen, stands

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free from this mass though arched towards it (fig. 10A).

THE LARVA

First Instar.—After three or more days a small space at the free end of the egg becomes glassy white. Soon a pair of tiny dark-tipped mandibles is visible through the thin shell and the faint outlines of body segments of the grub appear. Finally, the egg shell splits at this end and quite gradually slips down on either side and backward, thus exposing the glistening white footless grub, its jaws facing that upon which it is soon to feed. Its silvery threadlike tracheal system is now visible along the sides of the body. As the grub hatches, its body, already curved towards the abdomen of the tarantula, to which it is glued at its tail end, bends still more until, arch-like, its head touches the body of its host (fig. 10, B and C). Now sharp-pointed mandibles operate and the fleshy lower mouth parts are also applied to the skin of the tarantula so that a perforation is made, probably in part by the action of a dissolving fluid. Thus, the tiny *Pepsis* grub becomes firmly attached to the abdomen of the tarantula also by its mouth. One may now observe small movements at or behind the head, and as the grub sucks in the juices of the tarantula its body becomes colored. As it increases in size the egg shell shrivels to a dark pad near the end of the body.

The egg stage, as noted in a few instances for *Pepsis thisbe*, occupies from between four and five days, in quite warm weather, to ten days and some hours under cooler conditions, indoors.

Second Instar.—The wasp grub grows rapidly so that in a few days it undergoes the first moult or *ecdysis*. The old head capsule now cast off takes its place below and behind the new and larger head, and the old and wrinkled skin, including the lining of the tracheae, trails behind and adheres closely to the underside of the body. Soon the grub bends its head to the abdomen of the tarantula and resumes feeding through what appears to be a new perforation.

Third and Fourth Instars.—Moults 2 and 3 ushering, in larval instars 3 and 4, take place at the proper intervals. Except for the larger size there appear to be no other changes of note. There is the same type of finely pointed mandibles with denticles ranging down from one side of the tip (fig. 13A), the same smooth apparently naked skin, and the tracheae about as before. Two successively moulted larger head capsules, one behind the other, closely follow the first capsule beneath, and the attached moulted skins, thin and inconspicuous, underlie one another on the underside of the grub. Eyes are lacking as before. Feeding is still entirely by suction and the colored food contents may be seen churning in the body. The fully-fed fourth instar grub is large (fig. 11, 14). Naturally by now the abdomen of the tarantula has collapsed a bit at the feeding point of the infant wasp.

Fifth (Last) Instar.—With the fourth moult beginning the last instar some changes may be observed. The head, now pale brownish, is provided with a pair of stout three-toothed mandibles fitted for cutting and chewing, in keeping with a change in eating habits (fig. 12, 13B), the body is sculptured with delicate incised parallel lines or tiny creases encircling the segments, softening their brightness and somewhat obscuring the tracheal system. The lateral ridge is conspicuous for nearly the entire length of the body. Ten pairs of dark-rimmed, vertically oval spiracles are twice as large as in the preceding stage, and similarly distributed, as follows: One pair each on the second and third thoracic segments, and one pair each on the following eight abdominal segments. The pair on the third thoracic segment is very small. Four head capsules with accompanying skins adhere to the underside of the body. The semi-transparent tail end of the body beneath has a pair of granular patches.

Not for long now will the grub remain almost immovable on its host to suck in nourishment as in the earlier instars. It becomes relatively active, especially at the rather slender and muscular fore part. It easily cuts a hole into or enlarges the feeding puncture in the tarantula's abdomen, thrusts head and part of the thorax through this opening and feeds ravenously (figs. 15, 16). It bites through the rather stiff shell of the cephalothorax and reaches far into the bases of the hairy legs. The tarantula has expired, probably at the beginning of this attack. Usually no more than a few hollowed remains will attest its former existence. The wasp grub which, just after its last moult was loose-skinned, is filled out again and huge. It may attain a length of nearly two inches. Its skin is quite shining, for the fine folds have been stretched out by growth. Its final meal (?) is a dark finely chewed mass which has accumulated on its own broad underpart. These spider remains, however, may be partly incorporated in the cocoon which will soon be spun. The grub may now scrape or bite at its underside and even work its jaws when they are not in contact with any object.

DURATION OF LARVAL INSTARS

This particular *Pepsis* grub whose development we have been following has taken a long time to grow up: egg stage, 10 days and a few hours; first larval stage, about 3 days; second larval stage, 3¾ days; third larval stage, 4½ days; fourth larval stage, 6½ days; fifth larval stage, to just before spinning the cocoon, about 7 days; total, egg plus larval stadia, about 35 days. Under warmer conditions, and probably with different species of *Pepsis*, no more than

20 days may be required for these transformations, and, at least, in stadia 4 and 5 the former may sometimes exceed the latter in duration.

THE COCOON

To ensure the spinning of a well-formed cocoon I arrange cell walls within spinning distance of the grub. These walls consist of a narrow strip of corrugated carboard curved and placed on edge. A piece of clear glass is placed across these walls to function as a ceiling as well as a view window.

The grub first spins a delicate framework or support for the cocoon (fig. 17, *Pepsis mildei*). The silk issues from a transverse slit situated well forward on its protruding lower lip or *labium* (fig. 12,s). The grub reaches here and there, pushing into the remains of the tarantula and extending its fore part to fasten the silken threads to the walls and ceiling or to bite the strands together. This framework is now glassy white and shines like tinsel but in time these glistening threads lose their brightness. The grub spins while lying on its back or on its side and, when reversing its position in order to work on the other end of the structure, it executes a headfirst turn by means of slow undulations. The grub is quite as broad as, and stretches somewhat longer than the cocoon which is now taking form. It may reverse its position every half hour or less, laying down the silk, area by area, so that the structure acquires a pretty uniform thickness. It may be more than a day before the grub is hidden behind opaque walls. Then however, the cocoon is far from completed. We are informed by ticking sounds that the hidden worker is still busy, probably at the inner envelope. The cocoon, now stiff and leathery, is a long oval, nicely rounded and much wider at the head end; the tapering posterior may be slightly asymmetrical. It may be as much as 1⅞ inches in length.

This particular cocoon is whitish with a tint of tan and tends to be woolly. Cocoons formed underground are dirty gray or grayish brown, probably because soil is incorporated. The cocoon seems always to be spun so that the head end faces the filled-up tunnel.

Now the grub evacuates once for all the contents of its alimentary canal, this ***meconium*** forming a hard mass at the narrower end of the cocoon, which may be slightly discolored thereby.

A completed cocoon shows a thin tough outer wall, the inner side of which shows closely spun golden strands. This surface is connected by many threads to the rather frail inner envelope. The latter may be less developed at the head end and terminates as a skirt or curtain close to the narrower end of the cocoon. The interior wall of this envelope is varnished a rich brown color.

THE GRUB IN THE COCOON

The grub is now a good deal smaller than before it commenced spinning because it has expended so much of the body contents in making the cocoon and in voiding the alimentary canal. It has turned amber yellow, looks greasy and is deeply segmented. If it has formed the cocoon early in the summer it may turn into a pupa in a few weeks and hatch the same year. But if it is a late summer or autumn brood it is most probably destined to remain in the cocoon as a grub until the following late spring or summer. The enclosed grub can wriggle vigorously.

THE PUPA

When very near pupation the grub lies extended and quiet. When the change does come, the front part of the head capsule splits along the middle line, this break continuing through the back of the head and along the skin of the fore part of the grub's back and the very delicate glassy whitish pupa forces its way out of the shrivelling larval skin, which becomes wadded to the rear. The pupa (fig. 19) measures up to about 1½ inches in length. It soon loses its rather humped form and turns creamy yellow but with the large compound eyes showing grayish blue through the thin integument and giving it the alive look. The slender antennae, the legs and the stubby wing pads, rest freely but immovably against or close to the body. Two pairs of spicules adorn the upper part of the head in front, with others on the basal part of the antennae. On the middle of the dorsum of the thorax is a closely paired spinose process. Laterally on each side of segments 2 to 5 of the abdomen is a forked process, the first of which is in one plane, the others much curved. On each side of segment 6 is a smaller slightly knobbed process. On the back of segment 2 are many little pustule-like spines, while along the free edge of the back of this and of the three segments following are little spines that become larger and clustered mesad. The lateral abdominal processes are very conspicuous in the Pompilidae, and enable the pupa to fit more snugly in the smooth-walled cocoon.

The pupal stage may occupy several weeks.

EMERGENCE FROM THE COCOON

The pupa darkens as the enclosed wasp developes. When fully formed the wasp frees itself from the pupal skin, the wings quickly attaining their full length. *Pepsis* rests and gains more hardness of body. In good time the wasp may be heard buzzing and the familiar *Pepsis* odor permeates faintly through the seemingly impervious cocoon. A sharp scraping noise is audible at the broader end of the cocoon. Then a strong black mandible appears cutting through the thin tough wall (fig. 23), first on one side and then on the other, until this end of the cocoon is pushed off as a neat cap plus a sliver or two, and the wasp climbs out energetically (fig. 25).

The hatched cocoon contains the shrivelled

larval skin, the torn pupal skin, the larval meconium and some pellets voided by the emerging wasp.

On emerging from the cocoon the wasp digs its way out of the large cell, probably through the filled-in tunnel.

DURATION OF LIFE CYCLE

Our *Pepsis thisbe* has completed the cycle, from egg to hatching of the adult in 249 days, of which the cocoon period was 214 days. Another female wasp of this species required 350 days for its transformations. Where double brooded the shorter brood was estimated to require from about two to three months or more from egg to adult.

Adults may live for several months in captivity.

The habits of *Hemipepsis ustulata ochroptera* are very similar to its relatives, the *Pepsis* wasps, than which *Hemipepsis* seems lower in the scale of evolution.

The Material Studied

THE WASPS

Growing along a roadside ditch about half way up the golden slopes of Mount Diablo (elevation 3849 feet), some thirty miles east of San Francisco, is a colony of narrow-leaf milkweed (*Asclepias fascicularis*). Every summer its flower heads attract many insects the most conspicuous of which are the *Pepsis* wasps. Two species were thus observed, the larger widely-distributed *Pepsis thisbe* Lucas, up to about 1½ inches long, and the smaller *Pepsis pallidolimbata smithi* Hurd, hardly an inch in length, and of more restricted range. The male *Pepsis*, generally smaller, with slighter bodies than the females and with rather thickened antennae held straight out or slightly curved, range over the milkweed blossoms pausing to feed at nectar or searching for females, since it is here that mating may take place. During the heat of the day however, males often rest singly or in little groups on the flower heads and upper leaves that are in the shade of taller vegetation. The females, tremendous workers that they are, seem always to be desperately hungry. Some of them have frayed wings, perhaps a portion of an antenna or leg missing and bodies dusted with red soil, and are veterans that have fought and dug with sturdy vigor, while others recently from their cocoons are resplendent in their iridescent blue-green bodies and perfect wings. No need of a net to capture these giants; a carefully maneuvered jar will serve this purpose. So intent are they at securing nectar that you may gently interpose the hand between a flower and a wasp and she will crawl over, or rest on the hand in its search for food (fig. 2). On the ground beneath the plants one or two of these wasps, almost too decrepit to fly, are sipping at a puddle of water or trying to climb up the flower heads. The big females take wing with a deep hum that is sometimes accompanied by a ticking sound. They circle in the air and disappear.

In the foothill region of La Mesa, a few miles inland from the city of San Diego, *Pepsis chrysothemis* Lucas, *Pepsis mildei* Stål, *Pepsis mexicana* Lucas, *Hemipepsis ustulata ochroptera* Stål, and again *Pepsis thisbe* were studied. Among the flowers visited here by these wasps were the Milkweed (*Asclepias fascicularis*), Flat-top (*Eriogonum fasciculatum*), Sweet Fennel (*Foeniculum vulgare*), Toyon (*Photinia arbutifolia*), Goldenrod?, and Chaparral broom (*Baccharis sarathroides*); the shrub *Chrysosthamnus nauseosus* var. *gravicolius*[2], was studied in the Sierra Juarez mountains, Baja California.

THE TARANTULAS

The large bristly tarantulas commonly met with in California and which I have used in nearly all this work, belong to the genus *Aphonopelma* (fig. 3). I have determined none to species. Chamberlin (1940) described *Aphonopelma reversum* and *Aphonopelma eutylenum* from San Diego. Other species also occur in California. Despite their size they are not too readily identified. These *Aphonopelma* were collected on Mount Diablo, in Contra Costa County, and far to the south in San Diego County. Large female tarantulas as measured from the front of the cephalothorax to the end of the abdomen are about 1¾ inches in length, or fully 2 inches if we included the stout knuckle-like portion of the chelicerae that protrude beyond the head. The cephalothorax has a dense wool-like matting of pale hairs on the upper side while the legs and abdomen are covered with longer bristly, and with short hair. The underside is mostly dark and velvety. The eight small eyes are grouped on a low elevation on the head. From the position of these eyes it seems unlikely that the tarantula can see well, if at all, objects that are close before it on the ground. Speaking of the sight of tarantulas, Dr. Alexander Petrunkevitch, noted authority on spiders, says (1952, p. 52): "Their sight is poor, being limited to sensing a change in the intensity of light and to the perception of moving objects." But the tactile sense of the tarantula is very well developed, and drafts and vibrations affect it. The hollow poison-bearing fangs are hinged, somewhat after the manner of the blade of a jacknife, forward to the underside of the stout basal segment of the chelicerae. The tarantula seizes its prey with its chelicerae and other fore appendages, injects a poison, kneads and crushes its victim with the chelicerae while sucking the juices with its mouth. The pedipalps are the second of two pairs of appendages of the head. In the female tarantula

[2]Determined by Mrs. Ethel Bailey Higgins, Botanist at the San Diego Museum of Natural History.

they much resemble, in an abbreviated way, the four pairs of legs which follow. In the male however, the end portion of the pedipalps are modified into organs to transfer the seminal fluid to the female. The hair on the last segment of the legs beneath forms a sort of cushion enabling the tarantula to climb smooth vertical surfaces. Two pairs of spinning organs or spinnerets are situated on the underside very near the rear end of the abdomen.

The full-grown male tarantula differs greatly in appearance from the female. It has a somewhat larger cephalothorax, a small abdomen and much longer legs that may expand between five or six inches.

The common *Aphonopelma* tarantula lives in holes of its own making, or in old rodent burrows and large cracks in the soil. In some cases it is found beneath stones. It often occurs in small loose colonies, the members of which may not be in the same stage of development, as shown by the different diameter of the burrows in the colony. Several tarantulas in the laboratory were observed tediously lengthening the very short holes I had made for them along the glass side of each terrarium partly full of soil. A tarantula enters the hole headfirst to loosen the soil at the bottom with its chelicerae, and perhaps also with its pedipalps. Then it backs to the surface, turns about, backs in and applies the end of the abdomen to this loosened soil which it now webs over with silk from its spinnerets. Again leaving the burrow it re-enters headfirst, seizes the lump of cohering soil with its fore appendages, backs out with the load and deposits it at the surface. A burrow may be 8 inches or more in depth, though often much less. The gradient is usually steep. Typically the entrance is round to slightly oblong, with a diameter up to about 1½ inches. It has a very smooth look, for it is lined with silk to a short way within. Moreover, the tarantula's moving about at this entrance during the night helps mould this smoothness. A webbed-over entrance (fig. 1) shows positively that the tarantula is at home. Feebly-webbed balls of soil, as described, may more or less encircle the mouth of the burrow because the spider has been digging deeper or opening its winter quarters. And regularly cast out of its burrow are its moult skins, empty egg sac, and the remains of prey, such as sowbugs (Crustacea), and, very commonly on Mount Diablo, tenebrionid beetles of the genus *Coniotus*. Near San Diego, were recognized the remains of the golden-haired tenebrionid, *Cratidus osculans* (Leconte) about the burrows of *Aphonopelma*.

The smooth dark silky gray *Brachythele longitarsis* Simon is a smaller but more aggressive tarantula than *Aphonopelma*. It often lives in small colonies, digging steep holes that are sometimes nearly two feet in depth. These holes may be silk lined for a short distance, as well as silked over at the entrance. These tarantulas were collected chiefly in the San Francisco Bay region.

FIG. 1. Webbed-over burrow of an *Aphonopelma* tarantula. Mt. Diablo.
FIG. 2. *Pepsis thisbe* feeding at the flowers of milkweed. Mt. Diablo.

WASP VERSUS SPIDER

To witness a battle between wasp and tarantula in the open ground is too often a matter of good fortune. It is true that the wasp entices or drives the tarantula out of its burrow to battle above ground. Or, the encounter may take place in some deserted ground squirrel or other burrow deep among bounders. One may, however, stage such encounters in the laboratory,

through the use of such an arena as, preferably, a large glass-sided box, for example, an aquarium or a large jar with at least two flat sides. These receptacles should be filled to a depth of at least six inches with slightly moist soil well tamped down. The receptacle should be covered with a fine metal screen. Commonly, two receptacles are used in each complete experiment, the first receptacle constitutes the arena where the attack or battle takes place, while the second receptacle has been prepared for the burial ground. The transfer from one receptacle to the other, of wasp and paralyzed spider is easily done, though occasionally the wasp protests.

Petrunkevitch (1926), while in Puerto Rico, observed over 200 encounters in the laboratory between the tarantula *Cyrtopholis portoricae* Chamberlin and the wasp *Pepsis marginata* Palisot de Beauvois, not one of which was injurious to *Pepsis*, of which only three individuals were used in the experiments. He does not record the early development of the wasp beyond the egg that is laid on the abdomen of the tarantula, which is closely packed with soil underground. In another paper, Dr. Petrunkevitch (1952) discusses the odd relationship between the tarantula and the giant wasp *Pepsis:* "Although the tarantula can easily kill Pepsis, one species permits the wasp to sting it and lay an egg on its body". Dr. Petrunkevitch's observations and mine are sometimes in disagreement, as discussed subsequently.

FIG. 3. A large female *Aphonopelma*, abdomen elevated in annoyance, faces the camera. About natural size. Photo from kodachrome by E. S. Ross.

Lee Passmore (1936) shows his excellent photographs with description of a partial life history study on *Pepsis mildei* (reported as *Pepsis formosa*) including the attack by the wasp on an *Aphonopelma* tarantula.

The reader who is further interested in the biology of these wasps is referred to Dr. Paul D. Hurd's fine monograph (Hurd, 1952, pp. 265–270) and to Hurd (1948).

Pepsis thisbe Lucas

Pepsis thisbe Lucas 1895 (1894), Berlin. Ent. Zeitschr. 39: 732, 733, 737. ♂ ♀

This is a large tarantula hawk. The body is black with fine appressed hair or *tomentum* reflecting blue-green or purplish. The dark reddish brown wings have a dusky submarginal band.

On Mount Diablo one morning in mid July, males were observed swinging low over the slopes that were clothed with tarweed (*Hemizonia*), small *Eriogonum*, wild oats and other plants, or they flew about taller vegetation. Females were also about, alighting on the slopes and searching for tarantula burrows, pushing their way sturdily between the smaller weeds or travelling more rapidly in the open spaces. Thus a female might travel many yards, entering burrows in her path and then flying to another spot. Sometimes she re-examined a burrow. The burrows however, did not suggest those of tarantulas, and it seemed to me that her search was carried on in a rather hit-and-miss fashion, with no tarantulas brought to light. But perseverance is the essence of success in the hunting wasps.

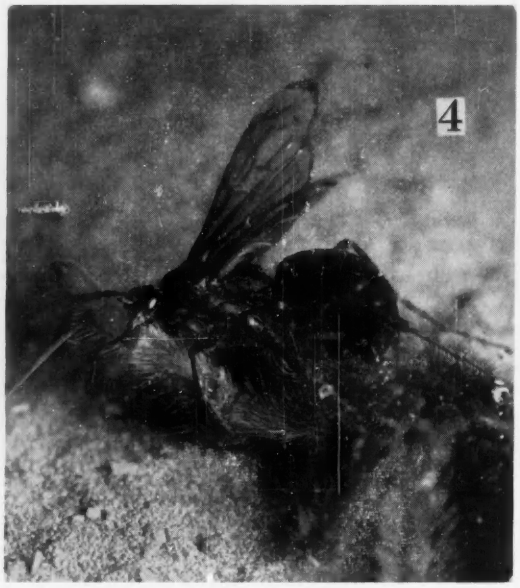


FIG. 4. A close up of *Pepsis mildei* stinging the overturned tarantula. The wasp is bracing herself by one of her victim's legs which she has seized at the base of the next-to-last segment. Somewhat enlarged. Photo by E. S. Ross.

Some very interesting results were obtained in the laboratory of our home in Danville, at the foot of Mount Diablo. An old *Pepsis thisbe* with tattered wings and two-thirds of one antenna missing was among several of these wasps cap-

tured July 24, 1952. She was designated as No. 3. When on the following morning she was examined as a possible worker, her frayed wings could not bear her aloft and she was judged too decrepit for use and released on the front lawn. But being still there a considerable time later she was recaptured. In her one month of captivity she parasitized ten tarantulas and proved the most satisfactory of my numerous *Pepsis* workers.

Here are some of her activities: August 16. After feeding *Pepsis* No. 3 with diluted honey placed on a piece of partly dried fig, I place her

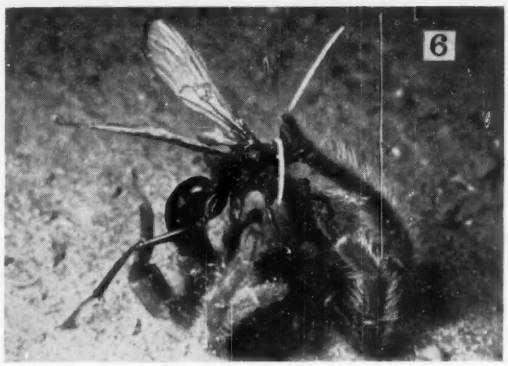


FIG. 5. Before the attack. *Pepsis mildei* following up the tarantula. Slightly reduced. Photo from kodachrome by E. S. Ross.

FIG. 6. The end of the fight. The victorious *Pepsis mildei* wasp has seized one of the paws of the overturned tarantula in her jaws and is administering the final sting in the cephalothorax of her victim. The pale orange antennae of the wasp are conspicuous. About natural size. Photo from kodachrome by E. S. Ross.

in an eight-gallon terrarium that is half full of soil. In this terrarium is a large fat-bodied female *Aphonopelma*. Upon the wasp's approach the tarantula faces its enemy and elevates its fore part in an attitude of defense. *Pepsis*, now about an inch away, readies herself to attack it by bringing her abdomen forward beneath her the more easily to use her sting. Her antennae move rapidly up and down. After some maneuvering she attacks the tarantula from the side. The tarantula grabs her with a lightning swiftness and turns on its back, but almost immediately it is very anxious to be rid of the wasp and attempts to push her away. The wasp, head facing the spider's abdomen, stems her fore part away and seizes one of the fourth legs near its tip in her jaws. She extends her abdomen in search for a place in which to insert her sting. Meanwhile the tarantula attempts to pierce the tough abdomen of *Pepsis* with its fangs. Finally the wasp finds the membrane between the two basal sections, the coxa and trochanter, of a fore leg and administers the paralyzing sting. The spider goes limp.

Pepsis now leaves the prostrate tarantula and drags herself, sometimes at a slight angle, over the soil, perhaps to rid herself of any of the tarantula's poison that may adhere to her body. (Note, however, that this instinctive cleansing procedure is sometimes employed when the tarantula has been subdued without a struggle!) The wasp appears unhurt. She approaches the tarantula and inserts her orange yellow tongue at the spider's mouth and sucks copiously. Soon she grasps the tarantula by the base of one of its second pair of legs on her side of the spider and attempts with little success to move its great bulk. She tries again, grasping in her strong mandibles the base of one of the third pair of legs on the far side of the spider's cephalothorax and with a strong pull turns it right over. An engineering feat!. The spider now rests underside to earth in orthodox position.[3]

After a bit, *Pepsis* finds the short "starter" hole I have made along the glass pane. Obligingly she continues the excavation and keeps her course along the glass. She loosens the soil with her stout mandibles and, with terrier-like strokes of her fore paws, she throws the soil behind and under her as she backs out of the burrow, and backing inches beyond the entrance, she scatters the load. A large lump of soil is carried in her manidibles, the fore legs taking over for a final fling at or near the surface. (Soil may also be "dragged" out, the chin or lower portion of the head assisting here, and, particularly if the gradient were steep, we might see *Pepsis*, with a heavy load of soil, curve her abdomen downward so that its point pushes again and again into the floor of the tunnel, with a slightly forward movement, thus assisting her backwards

[3]Petrunkevitch (1926: 379, 380, 387) observed *Pepsis marginata* similarly turning over its tarantula prey, to quote on page 379: "The Pepsis comes off the tarantula to the ground, takes a leg in her mandibles and begins to drag the limp body, stemming her own legs with great force against the ground. It is heavy work. The tarantula must be several times heavier than Pepsis and the ground is rough. In one place Pepsis finds it too difficult to drag the tarantula. She takes another leg and turns the tarantula over on her belly."

progress.) But the digging proves easy, for by 3:08 p.m., or about three hours after the digging had commenced, the gently sloping tunnel, including the enlarged cell portion at the end, is about a foot long. Except for my occasional scrutiny, the digging by *Pepsis* remains in the dark, with a piece of cardboard placed next to the glass pane.

At 3:14 p.m. the wasp is sweeping the floor of the cell with an extraordinary rapid movement of the forelegs which move in almost a haze of speed so different from the sturdy strokes she uses in excavating. Now she turns around and goes to the surface of the ground, grabs the tarantula by a second leg, pulls it a little, lets go, grasps the tarantula by the base of one of its fourth legs, gives a little pull, lets go her burden and descends into the tunnel to do a little rough sweeping, backs to the surface, seizes the spider by the base of a fourth leg, and lodges it broadside at the mouth of the tunnel. Then following several visits to the tarantula, she goes to, or near the cell, for all the world like taking measurements to fit the burrow to the spider! Finally, *Pepsis* with many pushings and pullings directs the spider's abdomen so that it plugs the entrance to the tunnel. After a little more arranging below, she seizes the tarantula by a spinneret and backs with it by stages down the tunnel and into the cell. The tarantula slips down best in this position because the limp legs follow. The tarantula lies on its side. Now follows an interesting procedure. Looking closely at the cell with the aid of a flashlight and a reading glass, I see the wasp like a bareback rider, astride the tarantula's abdomen rapidly scraping now with one foreleg, now with the other the general area of the abdomen where her egg is to be glued. She employs the inner side of the fore tibiae and perhaps the adjacent portion of the tarsi to brush off, more or less, the velvety layer of fine hair so that the egg will be more firmly glued to the skin. Her head and antennae soon become dusty with this fine hair. At the same time she kneads with her jaws the spider's abdomen basad of her scraping feet while she prods the portion near the caudal extremity with the end of her abdomen. Following these few minutes of work *Pepsis* advances along the tarantula's body so that she is head to head with it. With the tip of her abdomen, the sting being retracted, she does some vigorous back-and forth scraping. This soon ceases and the delicate operation of laying the egg follows. From the extremity of her well-extended abdomen the sting is extruded and raised clear of the spider's abdomen, and the shining white egg issues from between a pair of partly membraneous sclerites that press against the spider's abdomen and are then withdrawn into the wasp's body. The egg has been glued, thicker end down, at a little before the middle length of the abdomen.

FIG. 7. An *Aphonopelma* tarantula collapsing on its feet as *Pepsis mildei* stings it. About natural size.

Now *Pepsis*, without looking back, walks out of the cell. She seizes a large lump of soil in her jaws, backs in a little way and with her forefeet passes it beneath her body so that it forms the first and a large part of the cell plug. She scrapes down more soil, throwing this behind her (Plate V, Fig. 9), pushing it home with alternate strokes of her hind legs, and tamping it thoroughly with the point of her abdomen. At 5:05 p.m. the burrow is entirely filled, although *Pepsis* is still brushing dirt here and there.

Her progeny successfully completed its development so that on June 17, 1953, a fine female *Pepsis* issued from the cocoon.

August 9, 1952. 2:07 p.m. *Pepsis* No. 3 quickly attacks a formidable black male tarantula that spans about five inches. They break loose. There is another struggle and the spider now on its back loses a hind leg, the appendage breaking off neatly between coxa and trochanter. But the wasp grabs the other hindmost leg near its extremity as a sort of brace. Meanwhile the tarantula tries to push *Pepsis* away and to pierce the abdominal armor of its enemy, but time and again the long fangs slip down harmlessly on either side of the tough plates. The spider is stung in the region of the chelicerae.

Pepsis, quite uninjured, now cleans herself, using her tongue and tarsal comb for this purpose. She examines the detached and twitching spider's leg, sucks the nearly transparent blood oozing from the broken end and does the same at the stub.

It is only on the third day that *Pepsis* has completed and stored her burrow. It is of ample bore to accomodate the very large spider.

August 14, 1952. Here we see how *Pepsis* "warms up" for the attack on the medium large tarantula. The wasp holds herself tensely, antennae moving rapidly up and down before her,

while her abdomen with the segments telescoping in and out very quickly, shows that she is excited and breathing much faster than usual. She successfully attacks the underside of the tarantula, which loses a leg in the struggle.

August 18, 1952. *Pepsis* No. 3. Despite an easy victory over the tarantula, *Pepsis* seems to need a good cleaning. She drags herself along the ground and pushes the lower part of her head in the fine sandy soil so that her mouth and lower cheeks become very dusty. She cleans her antennae, passes the fore tibiae and tarsi between jaws and clypeus and rubs the top of her head on either side with the fore tibiae.

August 22, 1952. 2:45 p.m. *Pepsis* No. 3. The old wasp meets her fate. She follows the large retreating female tarantula and attempts to seize the end of its abdomen. The annoyed spider, its body well elevated, scrapes fine hair in the direction of the wasp, and the spinnerets produce quantities of silk. Neither of these defenses deter *Pepsis* from attacking from the side. The tarantula seizes her with a lightning swiftness, using all its legs in a mighty hug, and turns on its back. Somehow the powerful spider has gripped *Pepsis* crosswise and back to itself, and immediately it crunches at the base of the wings. The wasp is quite helpless in this vice-like grip; even her extensile abdomen with its long sting cannot reach a vulnerable spot. This impasse continues for a minute or more when, with a couple of appliances I loosen the leghold of the tarantula so that *Pepsis* is freed. Nothing daunted by this experience, she immediately and successfully attacks her still prostrate opponent. But the wasp has been injured and runs about in a rather tottering manner and one of her middle legs is stiffened. But soon she appears nearly as capable as before, although requiring about five hours to complete her burrow. The spider is stored as usual.

But the next day the old wasp is quite feeble and instead of running high on her legs she creeps about in a circle. She does not react when I wave my finger before her. But *Pepsis* wasps do not die easily and another day passes before she has made her last feeble movements (fig. 26).

I cut open her very hard-shelled abdomen and find at least ten eggs within her reproductive organs. Four eggs are of good size, and the largest of these is about four millimeters long and probably nearly mature.

Pepsis thisbe No. 10. La Mesa, October 9, 1952. A rather decrepit old female wasp, is found under our fig tree, where she has become intoxicated from feeding on its semi-fermented fruit.[4] Nevertheless, she easily deals with a large *Aphonopelma* and parasitizes two more tarantulas before her natural death on October 23.

Pepsis thisbe No. 2. From Mount Diablo. The wasp disturbs a large tarantula which makes off slowly, body elevated. But now the underside of its body comes in contact with a small bit of *Ascelpias* plant. This it grasps with a lightning-like thrust and bites. Evidently the very tense spider mistakes this weed for its wasp enemy. In overcoming this spider *Pepsis* suffers an injury to one of its forelegs. Nevertheless the spider is buried and parasitized.

Pepsis thisbe No. 51. From La Mesa, May 13, 1954. This rather small female was taken at the flowers of Flat-top (*Eriogonum fasciculatum*). During the 32 days of her captivity she parasitizes seven *Aphonopelma* tarantulas, one of these on

FIG. 8. An annoyed *Pepsis mildei* drags her tarantula over an old hat. Less than natural size.

two separate occasions. She gives special care in closing her cell, biting out the soil and tamping it so that it will not spill into the cell. Not one of the irascible type, she appears to show no concern when I take up her precious prey she has just paralyzed. She successfully attacks both large and small *Aphonopelma* tarantulas. In one of her spider burials the collapse of the walls of the cell covers her egg beyond survival. I remove the earth-covered spider, brush off the shrivelling *Pepsis* egg., blow the body clean and present it belly up to the wasp. Quite unexcitedly she examines the prostrate spider and deliberately stings it at the base of a leg. Within two hours it is stored in a freshly dug hole. A good cocoon is produced.

Pepsis No. 51. I introduce this wasp in a terrarium at the bottom of which in its spacious self-dug burrow is a small tarantula. The spider is overcome underground, not appearing at all at the surface. A cocoon is formed.

[4]Passmore (1936) has observed *Pepsis* wasps "fall to the ground as though intoxicated after partaking too freely of the fig juice."

Pepsis No. 51. June 12. *Pepsis* becomes interested in the young *Aphonopelma* that faces her from within the open half-pint jar. She buzzes, approaches and retreats before it and finally walks under the spider, looks upward and backs out. The spider high on its legs, is evidently frightened. It soon walks out of the jar and allows *Pepsis* to crawl crosswise upon it, to grab a leg and to attempt to sting it beneath. Although the seized leg snaps off, *Pepsis* does not seize another leg but ranges herself in front of the "frozen" spider and stings it effectively in the mouth region. While there has been no tussle, *Pepsis* scrapes the lower part of her head and thorax in the dirt and then brushes herself off.

In the two cases observed, from the laying of the egg by this *Pepsis thisbe* up to the beginning of spinning the cocoon, 24 and 29 days respectively, were required. The six cocoons measured 33 mm, 36 X 14.5, 36.5 X 13.5, 38 X 15, 47 X 19.5, and 48 X 19.5 mm. Two of these cocoons developed a white fungus within, and at least three of the remaining four transformed to pupae which should have produced adults within two or more weeks, had I not interfered with them.

After thirty-two days of captivity, I give *Pepsi* her liberty. I open the container and alertly she produces the brisk repeated buzz so commonly used when *Pepsis* discovers a tarantula! For the moment she seems unable to fly. She steps onto the flower head of a Flat-top plant (*Eriogonum fasciculatum*) that I approach, and feeds with an eagerness that belies the adequacy of the food that I have given here these past several weeks.

Pepsis pallidolimbata smithi Hurd

Pepsis pallidolimbata smithi Hurd, 1948. California Univ. Publ. Ent. 8: 126, 142–143. ♂♀.

A rather small species with reddish brown wings. It appears to have the same habits as the larger *Pepsis*. She preys upon young *Aphonopelma* trantulas. In one instance when this wasp was placed in a terrarium with a large tarantula, the latter rushed upon her and inflicted a fatal wound by puncturing her abdomen.

Wasps Nos. 4 and 5, of August 1952, readily overcame and parasitized *Aphonopelma* tarantulas an inch long. The two cocoons that were produced resemble those of *Pepsis thisbe*, except for their smaller size and perhaps their slightly slimmer proportions. Each of these two cocoons produced a female wasp sometime during July, 1953.

Pepsis chrysothemis Lucas

Pepsis chrysothemis Lucas, 1895 (1894), Berlin. Ent. Zeitschr., 39: 731, 739. ♂.

A medium large *Pepsis* with fiery red wings and the prothorax rather hairy. From La Mesa, Nos. 20 and 21, late May, 1953. Generally loath to attack large *Aphonopelma* tarantulas.

No. 20 is the smaller of the two wasps. I walk her out of a jar that I hold just before a tarantula which is an inch long and thus not as long as the wasp. *Pepsis* almost leisurely climbs upon its cephalothorax and curves her abdomen forward beneath it. Only then does the tarantula spring into action. But it does not seize *Pepsis* which, easily keeping her distance stings it effectively beneath. Then the wasp follows the usual procedure.

Wasp No. 21 is a rather large specimen. She is at first placed in a terrarium with the smaller female of her own species and a rather small *Hemipepsis ustulata ochroptera*. She savagely attacks her own kind and attempts to sting her. The smaller *Pepsis* takes refuge beneath a clod. There is no serious hostility between *Pepsis* and *Hemipepsis*.

The three *Pepsis crysothemis* secured, hatched in August of the same year. The cocoons had their inner envelopes thin like tissue paper and with none of the intervening golden floss, as occurs in *Pepsis thisbe* to the contrary.

Pepsis mexicana Lucas

Pepsis mexicana Lucas, 1895 (1894), Berlin. Ent. Zeitschr. 39: 560, 561, 566. ♂♀.

An inch-long blackish winged wasp. From La Mesa, early August of 1953 and 1954. Found at the flowers of *Eriogonum fasciculatum*, and on the wing.

EXPLANATION OF FIGURES 9–13

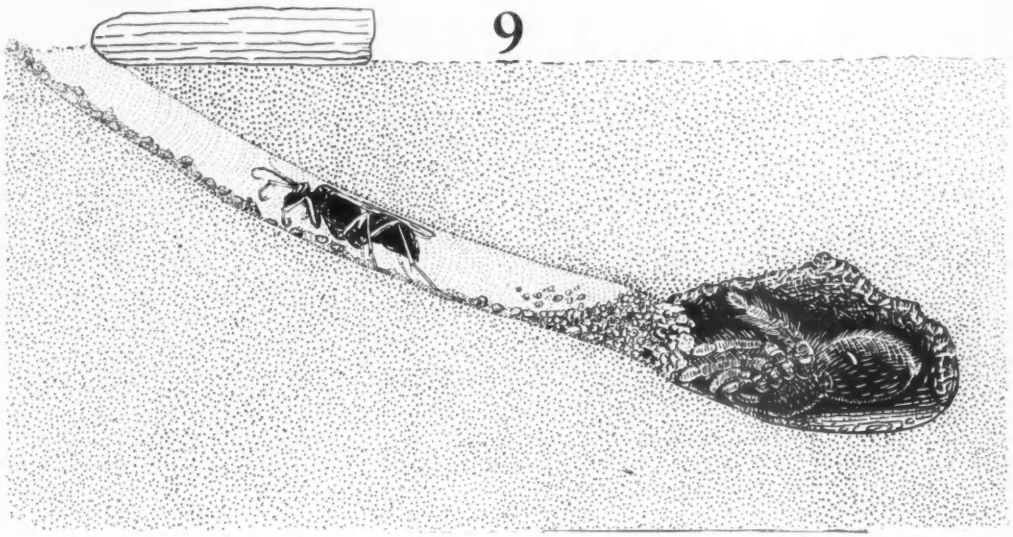
FIG. 9. *Pepsis thisbe* has stored the paralyzed *Aphonopelma* tarantula in the cell. The wasp's egg may be seen attached to the abdomen of the spider. The wasp (inserted in the drawing) has plugged up the cell and is now filling up the tunnel by throwing back soil with the terrier-like action of her forefeet. The tunnel has been somewhat conventionalized, and the flat stone used by the author to cover the upper edge of the "starter" burrow, is seen in position. Reduced.

FIG. 10. *Pepsis thisbe*. Egg A, and two larvae B and C, in the process of hatching, on the abdomen of the Tarantula. E, egg shell. The egg is about 4.5 mm. long.

FIG. 11. *Pepsis thisbe*. Larva, fourth or penultimate instar, feeding by suction on the abdomen of the tarantula. In this side view may be seen the moulted heads 1, 2 and 3, with their adhering skins and the shrivelled eggshell, E. The grub or larva is about 15 mm. long. Slightly diagrammatic.

FIG. 12. *Pepsis thisbe*. Head of the grub in the last stage. Eyes are lacking, and what seem to be occupying the position of the eyes and adding "expression" to the face are the much aborted antennae. The broken parallel lines indicate the limits of that part of the manidibles behind the broadly bilobed labrum and a portion of the clypeus. The opening of the salivary duct is the somewhat raised transverse slit at S.

FIG. 13. *Pepsis thisbe*. A, mandible of a fourth stage grub. The mandibles of this and the earlier instars function as scrapers or puncturers, and are heavily sclerotized only at the apical portion; B, mandible of last stage grub. They are fitted for chewing and cutting and are heavily sclerotized throughout.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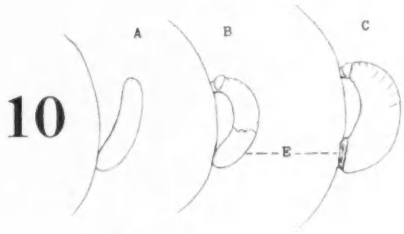
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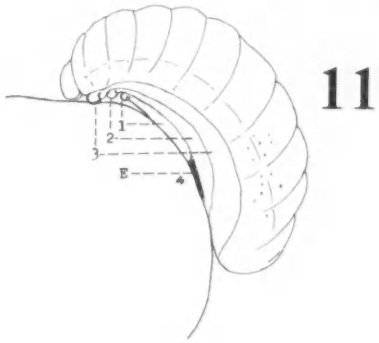
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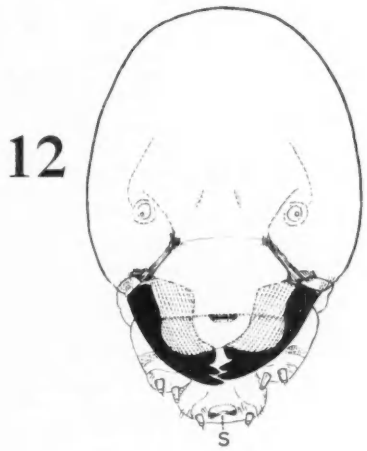
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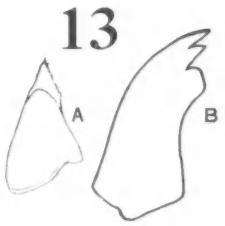

13
A
B

Pepsis mexicana No. 52. An inch-long *Aphonopelma* tarantula is placed in the terrarium with her but produces no response. Then a second tarantula similar in size and appearance is substituted. It is acceptable to *Pepsis*. But the little tarantula seems fully aware of its peril and makes a tremendous rush up the glass side and to the screened cover of the terrarium. It has to be pushed down constantly to the now thoroughly aroused wasp. She flies at the spider and with wings elevated, attempts to seize a leg. But now the tarantula remains quietly on the ground so that *Pepsis* is allowed to crawl on its back, to seize a leg and to sting it beneath in a vital place. The spider now rests on its back. Although there has been no struggle, the wasp scrapes herself. She quickly digs a burrow, stores it, refills and smoothes it over so as to hide any trace of its presence.

A month later I dig up the burrow. It is only four inches long and one and one/fourth inches deep. The spacious cell has the remains of the spider's fore part towards its entrance end. The cocoon is an inch long and its broad end is likewise toward the entrance end of the cell. The tail end of the cocoon is very narrow.

This wasp takes little interst in tarantulas that are obviously too large. Neither does she attack (in a rather brief exposure) the fierce though small and silky-haired *Brachythele longitarsis* tarantula that I have brought down from the north.

Pepsis mexicana No. 52. La Mesa, August 5, 1952. An experiment with three female *Pepsis mexicana* and several *Lycosa*, or wolf spiders. These arachnids are an inch or a little more in length of body and have rather long legs. The body has some pale stripes. They typify speed ferocity and alertness among spiders. They were found living in holes well up the slopes of Mount Diablo. These holes could be mistaken for those of our tarantulas, but in general, they are more webby about the mouth and sometimes there is an indication of a low turret there, but with no accumulation of soil about the opening.

In turn, each *Pepsis mexicana* is placed in a terrarium with one of these spiders. At first and very briefly one of these wasps may show a slight interest in *Lycosa*. But the reaction of *Lycosa* towards *Pepsis* is very vigorous. For a moment it stands at defense, then makes a lightning dash at the inoffensive wasp, seizes and bites her but quickly lets go. The wasp is quite upset but apparently unhurt. It avoids the spider now standing tall and alert. *Lycosa* apparently sees better and makes quicker turns than does the *Aphonopelma* tarantula. It may attack again when the wasp inadvertently approaches it, try to fight at long range or turn on its back, outspread and threatening. Even the big *Pepsis thisbe* is discomforted by its vigorous behavior. But *Lycosa* spiders do have their enemies among the pompilid wasps.

Eventually, one of these three wasps accepts her natural prey, a small *Aphonopelma* tarantula. She stings and buries it in her rather shallow tunnel.

Pepsis mildei Stål

Pepsis mildei Stål, 1957. Öfvers. Kongl. Vetensk. Akad. Forh. 14: 64.

La Mesa, November 18, 1952. A fine bright day. I follow a large *Pepsis* with orange antennae. She runs over the stony ground, peering into, or entering rodent holes, to emerge from the same or a connecting burrow. During the irregularly disposed quest, covering several hundred feet and occupying about fifty minutes, she makes but two short flights. Finally, she enters a mouse-size burrow and stays within for several minutes. She emerges for a little while and re-enters. After waiting several minutes I dig at the burrow and almost immediately see the waving orange antennae. The wasp is maneuvered into a jar. Two inches further down the hole is a large limp *Aphonopelma* with a rather thin abdomen. It has been overcome underground.

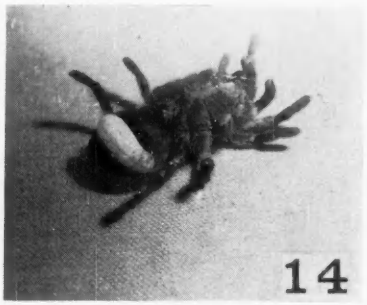
Placed in a terrarium, *Pepsis* turns the tarantula over on its belly, does some digging but does not bury her prey. After feeding at a fig daubed with honey and water she thoroughly scrapes her face and mouth in the loose dirt, then cleans them with the forefeet and passes her fore tarsi through half opened jaws. Finally, she rubs one mid tibia and tarsus and both hind tibiae and tarsi together, meanwhile balancing herself on the other three legs.

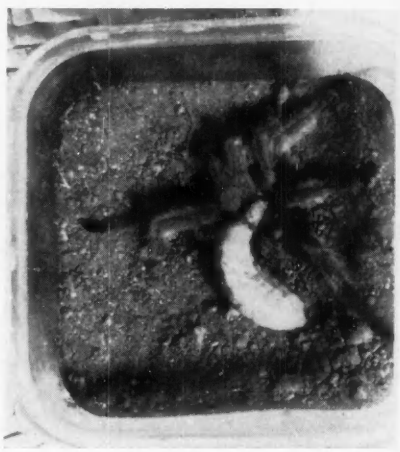
She lives 29 days and parasitizes no tarantulas.

Pepsis mildei No. 40. La Mesa, August 31, 1953. An old, ill-tempered wasp. She spends the first night under a piece of plant fiber in the terrarium and buzzes angrily and produces the *Pepsis* odor when I direct the beam of a flashlight towards her. The next afternoon she refuses successively two large tarantulas, but quickly stings a rather small one which she buries. She lives for 29 days.

Pepsis mildei No. 41. La Mesa, October 1, 1953. A fearless insect. Now she confronts a good-sized tarantula that rests rather cramped in an open half pint jar. She finally walks in and during the lively struggle that ensues the spider loses a hind leg and is stung to immobility, still in the small jar. *Pepsis* feeds at the spider's wound, and at 5:00 p.m., commences digging at the "starter" hole. It is difficult digging. From time to time she examines her prey and moves it a little. When I run my finger along the glass pane while she is above ground she dashes towards my finger in a very alert manner. She is still at work when I go to bed at 2:30 a.m., October 2. Many hours later the spider is buried, but the burrow remains only partly filled.

Pepsis mildei No. 42. From the Sierra Juarez





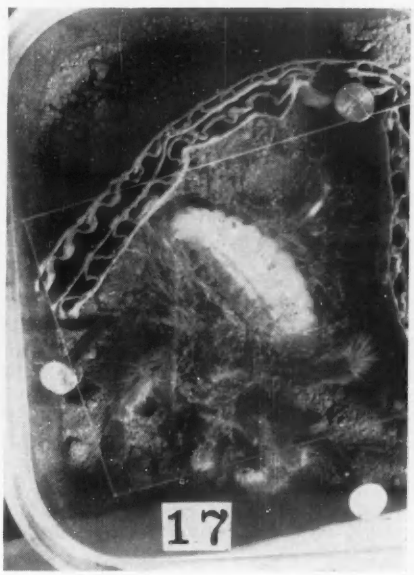


FIG. 14. *Pepsis thisbe*. Larva, fourth instar, feeding on the abdomen of the tarantula. About two-thirds natural size.

FIG. 15. *Pepsis mildei*. Larva, fifth instar, feeding at the fore part of the tarantula. About two-thirds natural size.

FIG. 16. *Pepsis mildei*. Larva, fifth instar, feeding at the fore part of the tarantula. About two-thirds natural size.

FIG. 17. *Pepsis mildei*. Larva, under a plate of glass, beginning to spin its cocoon. About X¾.

Mountains, Baja California, Mexico. On October 22, 1953, at 12:48 p.m., she is presented with a large female tarantula parasitized some days ago by another *Pepsis* but now bearing no egg. She stings the helpless spider in the membrane between the base of a pedipalp and of the first pair of legs on one side. Then she cleans herself. When I seize the spider with a pair of tweezers the angry wasp holds on to her prize as I lift spider with wasp from one receptacle to place it in another. Her nest hole is sixteen inches long, as measured along three faces of the jar. The tarantula in the spacious cell has the abdomen pointing *toward* instead of the usual away-from-the-fill direction. The wasp's egg arises from the side and just before the middle length of the spider's abdomen. It hatches in eight and one-half days. But the doubly-stung tarantula dies before the wasp is half grown, and the grub in turn dies some days later.

This performance is repeated with another previously parasitized tarantula, the developing wasp grub perishing but the tarantula maintaining its paralyzed existence. And finally, *Pepsis* stings and parasitizes a tarantula that has already been *twice* parazitized, and here the grub dies, while at this date, over a month later, the tarantula remains alive.

Tarantulas paralyzed by *Pepsis* may remain in this feebly living condition for months, and for the most part die through desiccation, since they cannot feed.

November 2, 1953. Figure 7 shows *Pepsis mildei* No. 42 stinging a tarantula.

The single slightly woolly cocoon of *Pepsis mildei* that I obtained (fig. 18) was at first pinkish white. Later it turned to pale brown. It is stronger and thicker walled than those of the other *Pepsis* species studied, for it has several additional closely packed silk layers.

Mr. Harry A. Hill of San Diego, has kindly transmitted to me his interesting observations on a *Pepsis mildei* wasp with its paralyzed prey, an *Aphonopelma* tarantula, and which I put down here in condensed form. It was one evening near sunset in early September, a few years ago that he discovered this *Pepsis* dragging her prey over the ground. During the next four hours or more, and thus well into the darkness, she dragged her burden an estimated 130 feet over some quite rough terrain. The tarantula was finally dragged to a hole that the wasp had probably prepared previously and now set about enlarging. It was then half past nine in the evening, and a flashlight and a kerosene lamp were brought into use. *Pepsis* worked over the burrow, buried the tarantula and refilled the burrow. "Presently when all was done she flew off slowly into the night." An examination of the burial spot the next morning showed . . . "a rather large shallow hole, and over all clung the strong odor of a skunk. For this reason we have always thought that the marauder was a skunk."

Hemipepsis ustulata ochroptera Stål

Hemipepsis ustulata ochroptera Stål, 1857. Öfvers. Svenska Vetensk. Akad. Förh. 14: 64.

This rather dull black hairy wasp differs generally from *Pepsis* in its somewhat stouter antennae and legs and in its wing venation. It varies greatly in size; in the small series of females before me the range is from three-quarters of an inch to one and one-half inches long.

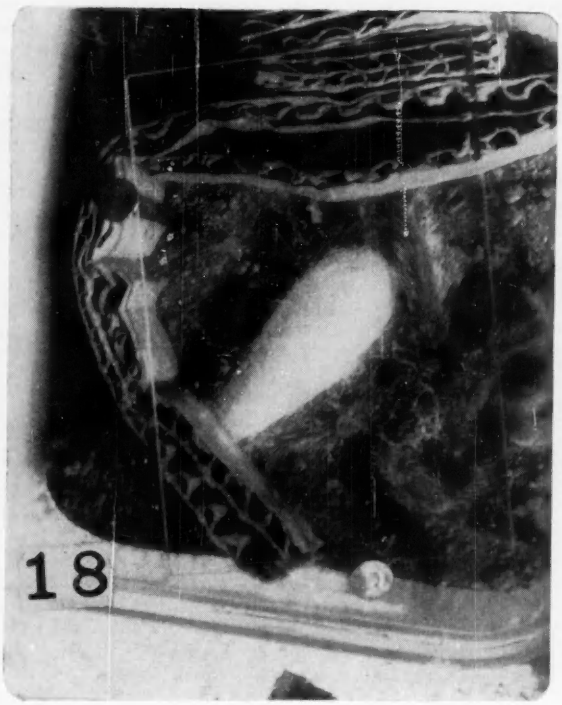
Hemipepsis ustulata ochroptera. Nos. 1–3. Menlo Park, San Mateo County, California, late summer of 1937. Several of these wasps were secured in the territory occupied by the silky *Brachythele longitarsis* tarantula. In the few experiments carried out in screened-over porcelain crocks, it was found that *Hemipepsis* parasitized these spiders. None was reared beyond the first larval stage.

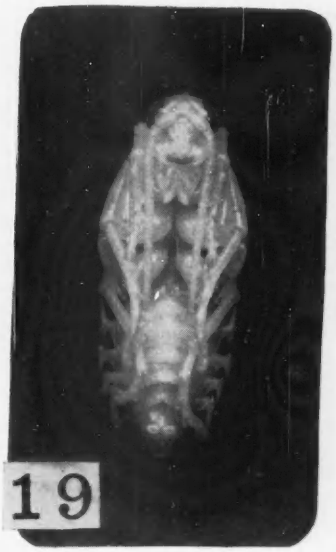
Dr. C. P. Smith (1908) who studied the systematics and habits of *Brachythele* and of other mygalomorph spiders in the above general area, reports *Pepsis* as an enemy of *Brachythele longitarsis*.

Hemipepsis ustulata ochroptera No. 11. La Mesa, October 12, 1952. A large female, in fresh condition, feeding at fermented figs.

December 17, I give *Hemipepsis* an *Aphonopelma* tarantula that has been parasitized by a *Pepsis* five months ago. It is capable of only feeble movements, its fangs being paralyzed and its abdomen shrivelled for lack of moisture. Nevertheless, *Hemipepsis* immediately climbs upon the spider, grasps one of its forelegs, and curves her abdomen to sting(?) it beneath the cephalothorax. She begins digging at the "starter" hole, but soon swerves off the glass pane. She appears to do less terrier-like digging but more hauling than do the *Pepsis* wasps. She holds the upcoming load of soil with incurved forefeet, the chin assisting, or, she takes up a lump of soil in her mandibles. At the steeper grades she humps up and pokes the end of her abdomen into the floor, repeatedly, as *Pepsis* may do, to help pull herself out of the tunnel. When at or near the surface, she brushes back the soil terrier-like.

She works far into the night. The shrivelled, rather unyielding spider is destined to give her a great deal of trouble, for the ten-inch-long tunnel is sharply bent in the rather small terrarium. She pulls at the spider's legs as if trying to limber them; she gets on her back under the spider; then she is right-side up again; at last drags her victim, tail first, into the two-inch-long cell. The wasp cleans her very dusty self. She refills the tunnel for a short distance, then leaves a long open space and then fills the outer two inches. The yellowish white egg, like that of *Pepsis*, is slightly curved and a little thicker at the tail or attached end. It is 4.25 mm. long. It is glued at one side of the ventral line and at





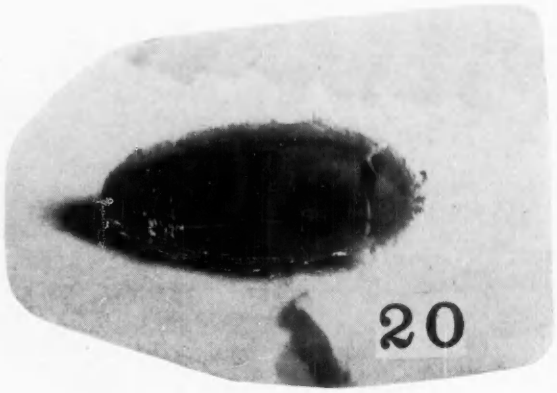


FIG. 18. *Pepsis mildei*. The cocoon completed. About natural size.

FIG. 19. *Pepsis thisbe*. Pupa, female. Ventral view. Length 34.5 mm.

FIG. 20. *Hemipepsis ustulata ochroptera*. "Hatched" cocoon. Length 35 mm.

FIG. 21. *Pepsis thisbe*. "Hatched" cocoon. Length about 42 mm.

the middle length of the spider's abdomen. Neither the tarantula nor the wasp grub survives.

One small and one rather large, active *Aphonopelma* tarantula are dealt with by this wasp on February 5 and 23, respectively. Altogether, at least four tarantulas were successfully parasitized.

On March 2, an encounter with a rather large brown female *Aphonopelma* proves fatal to the now stubby-winged *Hemipepsis*. In the fierce struggle the tarantula grips its enemy's head and thorax in its fangs. When I free the wasp she staggers about crazily, and while later recovering somewhat, dies the next day. She has lived 143 days in captivity. Her two sons and one daughter emerge the same year (1953). The fourth and largest cocoon contains a dead larva.

Hemipepsis ustulata ochroptera No. 15. Taken near La Mesa, August 31, 1953. This wasp is of medium size. On September 3, she parasitizes a fairly large *Aphonopelma* tarantula. On September 6, I place her in a gallon terrarium containing a *Brachythele longitarsis* tarantula in a large deep open burrow. The spider is a little over an inch long. *Hemipepsis* explores the burrow. Immediately there is a struggle and the tarantula rushes to the surface, followed by the wasp that manages to cling to it for awhile. Now the wasp extends her abdomen far forward beneath her and stings the spider at long range. This stinging is rather prolonged. The wasp turns the spider belly up. She spends over two hours working at the spacious tunnel of *Brachythele*. She does much filling in. The spider is buried and the wasp refills the tunnel, turning around often as if to inspect her work.

On September 23, I exhume *Brachythele* but find no trace of parasitism. The tarantula has recovered somewhat, for it makes a show at defense and extends its fangs a little. I place this enfeebled spider with its former conquerer. The wasp stings it, and after a number of attempts succeeds in turning it on its back. Then she gives it a long sting in the mouth region. She spends much time examining her prey and probes or rubs its abdomen with the tip of hers. After some hours the tarantula is buried. The cell has a view window, and we see the spider resting on its back, the wasp's egg glued near its lung slits. The egg hatches in between four and five days, but the much-stung spider soon dies and so does the grub, when half grown.

September 17–19, 1953. *Hemipepsis* No. 15 is placed with a huge though rather mild male *Aphonopelma*. The wasp seems perplexed by the tallness of the spider. She walks beneath it, reaches upwards and examines it from the side. When the giant settles on the ground, she climbs on its back and attempts to sting its underside, but is prevented by the close-set hairy legs. Finally, when the spider stands tall again she once more tries to get a grip and even flies up a little. She climbs part way up a hind leg (fig. 22) but gets no hold among the bristles and falls back. The annoyed spider lifts a leg as if to repel its tormentor, and when the latter ventures near the mouth region it seizes her in a vigorous hug, turns on its back and releases her. The wasp seems disconcerted, but some time later, when given another try, this ludicrous performance is repeated.

Among her later experiences *Hemipepsis* shuns a large female *Aphonopelma* but readily parasitizes two small ones of the same genus.

Some time later she attacks a large *Aphonopelma* that turns on its back and with paws brought close together desperately pushes away its enemy, the scene suggesting a juggling act! But *Hemipepsis* has seized a hairy paw, is able to use her sting effectively.

Finally *Hemipepsis* meets her fate. She battles with a very sturdy female tarantula that repels her in two encounters. Next day the wasp is quite feeble and on the following morning is dead. Examination shows a fluid exuding from between the terminal plates of her abdomen. Evidently the fangs of the spider had penetrated there. Life in captivity has lasted 63 days.

The cocoon of *Hemipepsis* differs from that of *Pepsis* in being a beautiful brown silk structure that is only slightly wider at the end from which the wasp will emerge. Its inner wall is varnished, but there is no inner cocoon. As with *Pepsis*, *the Hemipepsis* wasp cuts off a neat emergence cap (fig. 20).

RESUMÉ AND DISCUSSION

The five species of *Pepsis* wasps and the one species of *Hemipepsis* wasp studied were found to parasitize tarantulas of the genus *Aphonopelma*. In addition, the *Hemipepsis* wasp parasitizes the *Brachythele longitarsis* tarantula. It seems very probable that all these wasps will accept more than one species of the genus *Aphonopelma*.

The large species of *Pepsis* successfully attack and parasitize tarantulas ranging in size from quite small immature ones to fully grown specimens; the small *Pepsis* (*P. pallidolimbata smithi*, *P. mexicana*) appear unwilling, if not unable to

EXPLANATION OF FIGURES 22–24

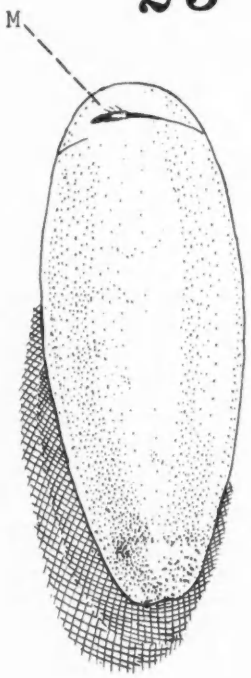
FIG. 22. *Hemipepsis ustulata orchroptera* attempting to climb up a leg of a male *Aphonopelma* tarantula in order to sting it. The wasp is slightly more than an inch long. From a photograph.

FIG. 23. *Pepsis thisbe*. Cocoon. The wasp is preparing to emerge, and one of her mandibles (M) is seen shearing off an emergence cap. Length of cocoon 40 mm.

FIG. 24. *Pepsis thisbe*. Fore part. To show how she cleans her antennae by passing them through the comb grip of the fore tibia and tarsus. For clarity, some spines have been omitted. Enlarged.

22

23



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cope with mature tarantulas, but do not hesitate to attack young ones of a size suited to the development of their own progeny.

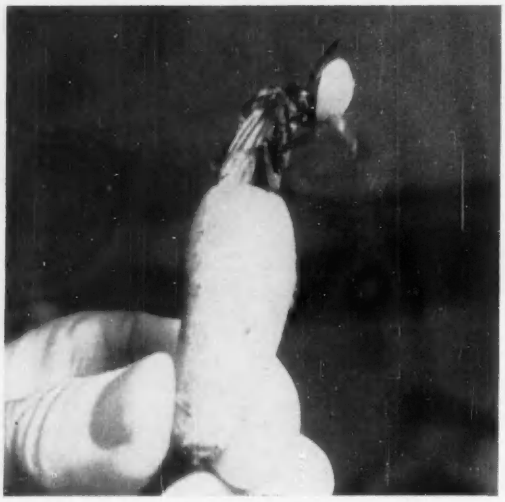

FIG. 25. The birth of a *Pepsis thisbe* wasp. Issuing from a cocoon held between the fingers. Slightly enlarged.

In the terrarium these wasps often hastily avoid the threatening spider, as if in fear; perhaps the spider is too large, or perhaps they are not physiologically ready to attack it. Usually there is some maneuvering before the battle; the tarantula, fore part raised, faces its enemy, occasionally striking at her; the wasp, not immediately closing in, bends her abdomen towards the tarantula, so as to use her sting more readily, and to try to sting its underside. The result of this approach may be the seemingly deadly embrace of the wasp by the spider, and the "crunching" of her wings by its fangs; the reverse may occur when the hard-shelled wasp pushes herself partly away from the spider, having already seized one of its legs (sometimes snapping it off) in its mandibles, to brace herself. During this brief but deadly struggle *Pepsis* has been probing with her sting for a membraneous place in which to deliver her paralyzing thrust.

But there may be considerable variation in the manner of the struggle. When first aware of its enemy the tarantula may rush upon her and endeavor to kill her; at the other extreme, baby tarantulas will rush away from the scene of danger. These spiders when on their backs are also adept at pushing away their enemy. Not infrequently the wasp, her abdomen curved far forward, is able to sting the tarantula at long range, and thus avoid a rough encounter. Or finally, the tarantula may become terror stricken and without a struggle permit the wasp to paralyze it.

Occasionally the wasp loses the battle with the tarantula, or both may be rendered *hors du combat*, as described by Kellogg (1908).

The wasp stings the tarantula in the membrane separating the basal segment or coxa of a leg from the next segment or trochanter, in the cephalothoracic membrane between the leg bases, and/or in the mouth region. All such stings are near to or at the large nerve ganglia.

These pepsine wasps will often accept a tarantula that has been previously paralyzed, even months before. Such a spider is now stung again, for the wasp follows her instinct, being quite incapable of knowing that the spider has already been sufficiently stung. The spider is then buried and parasitized, usually to no purpose.

The furious battles that may take place between two wasps over a tarantula which one of them has stung, may sometimes explain the more or less crippled condition, an antenna or leg portion

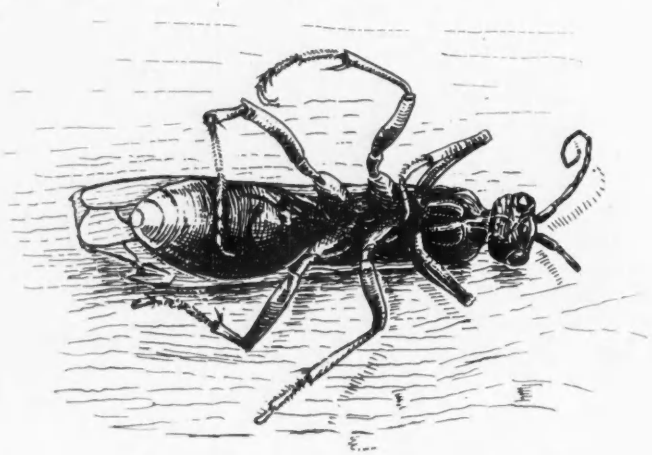

FIG. 26. *Pepsis thisbe*. Journey's end. Enlarged.

missing, of female wasps we often find afield. But at least in the laboratory the tarantulas are sometimes responsible for damage to the wasps that attack them. And perhaps the age-old habit among males of many organisms of fighting for the possession of the female may explain the occasionally imperfect condition of a male wasp.

Tarantula hawks (at least *Pepsis mildei*) often show great "courage" when molested by man in relation to their hard-won tarantula.

It is more usual, perhaps, for the wasp to dig a burrow or select a deserted animal burrow, or a crack in the earth as a nest-hole *before* she hunts for the tarantula. Thus, the tarantula stung where the wasp has found it, may be far from the wasp's nest-hole. Rau and Rau (1918, on pages 67–71) speak of a *Pepsis* wasp that dragged her immense spider victim for a hundred yards to finally disappear with it into a rodent hole. The observation by Mr. Harry A. Hill, quoted in the present paper under *Pepsis mildei*, also illustrates this point. Going farther a'field, the findings of Hingston (1925: 80–89) on the pompilid wasp *Cryptocheilus rubellus* Eversmann, near Baghdad, Mesopotamia, are of interest. This wasp attains a length of about an inch and a quarter. She seeks the burrows of a "Tarantula", evidently one of the mygalomorph spiders, ousts the giant from its burrow and pursues it. She is very wary in attacking her formidable adversary and seeks to sting it at long range or when it has sought a disadvantageous shelter. *Cryptocheilus* first may sting the spider in the abdomen, and thus crippled it becomes an easy prey. Then the wasp drags the paralyzed tarantula directly into a deep crack in the earth where it is stored. This retreat, which may be very distant from the scene of battle between wasp and spider, has evidently been selected by *Cryptocheilus* before she set out on her hunt for the spider.

Pepsis may utilize the burrow of the tarantula she has immobilized, as described by Passmore (1936).

Finally, *Pepsis* and *Hemipepsis* may overcome the tarantula underground, where the burrow or other cavity is roomy enough, and bury it there.

After the tarantula is overcome by *Pepsis* it usually rests upon its back. Usually before beginning her borrow *Pepsis* turns the tarantula over so that it rests belly to the ground. When the tarantula is stored in the cell it rests upon its side and almost invariably faces the entrance, now plugged up. My less complete observations on *Hemipepsis* seem to indicate that the tarantula is usually placed on its back and that it rests thus in the cell.

The burial of the tarantula by *Pepsis* and *Hemipepsis* as recorded in this paper differs markedly from that by *Pepsis marginata* with regard to the tarantula *Crytopholis portoricae* studied in Puerto Rico by Petrunkevitch (1926) as quoted from page 388: "She packs the soil until it so invests the body and legs of the tarantula that even in case of recovery the unfortunate creature could not move a single limb." The six species of pepsine wasps studied by me all dug firm commodious cells in which to store the tarantula. But two cells did collapse because the soil was too loose. There are however, certain Pompilidae, like *Tachypompilus*, that pack the soil over their spider victims after having glued their egg on the downturned body of the spider.

The mosaic vision of insects seems best suited for recognizing moving objects and, to supplement her eyesight, we see *Pepsis* constantly tap the ground or other objects with her antennae, which seem to be organs of smell as well as touch. She certainly has an adequate sense of distance and dimension. Working in her dark tunnel, she knows when it is deep enough and the cell sufficiently spacious. And she is aware of what part of the spider's abdomen is to be prepared for gluing her egg. She never seems to turn around and look at her egg but forthwith plugs up the cell.

In filling the burrow tarantula hawks do much tamping or pressing with the extremity of the abdomen. Then they often assume an upright position or partly suspend themselves to the wall by a leg. Tamping is widely used among digger wasps. In addition to tamping by *Pepsis*, the soil is thrown back with the forefeet and then also and to a less extent pushed back with the hind feet.

Finally, it must be remembered that the majority of these observations were carried on under artificial conditions, involving frequent disturbance, insufficient space, frustration, fright, anger, etc., which sometimes may have affected the behaviour of the wasps and the tarantulas differently than under natural conditions.

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THE MORPHOLOGY AND FUNCTIONAL ANATOMY OF THE MALE AND FEMALE REPRODUCTIVE SYSTEMS OF CIMEX LECTULARIUS L. (HETEROPTERA, CIMICIDAE)[1]

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INTRODUCTION

The unusual adaptations of the reproductive system found in the common bed bug, *Cimex lectularius* Linn., have engaged the attention of several investigators during the past years. The right genital clasper of the male is a highly modified claw-like structure which penetrates the intersegmental membrane adjacent to the organ of Berlese and thus carries the aedeagus into the latter. Since the organ of Berlese has no direct connection with the reproductive tract, the spermatozoa must pass from it into the body cavity and then penetrate a special lobe at the base of each lateral oviduct where they are stored. From here some migrate in the wall of the oviducts up to the ovary where they fertilize the developing ova. While insects generally produce eggs whether mating has occurred or not, the bed bug does not lay eggs until the female has mated. The manner in which insemination initiates complete egg formation is still not completely understood.

The peculiar mode of insemination of the bed bug is also found in many other species of Cimicidae and may be of general occurrence in this family. In Anthocoridae, a closely related family, certain genera reproduce in a manner very similar to the bed bug. Other families of Heteroptera such as Reduviidae, Miridae, and Nabidae likewise show special modifications of the way the sperm is stored in the female. In each of these families the spermatheca no longer serves for the retention of spermatozoa, and this function has been taken over by certain new organs.

Notwithstanding the previous studies various aspects of the male and female reproductive systems of *C. lectularius* merit a more intensive investigation. This study was therefore undertaken with a particular goal of providing additional information regarding the functioning of the reproductive organs as well as contributing to a more substantial basis for the interpretation of how the unusual features in the reproduction of cimicids and related heteropterans probably evolved.

LITERATURE

The literature concerning the reproduction of the bed bug prior to 1920 has been thoroughly reviewed by Cragg (1920) and therefore only the principal aspects of this literature will be considered here. It is felt that such a consideration may be of much value since Cragg's paper, as well as studies preceding it, appeared in journals not commonly available to entomologists.

The earliest observation on the unusual structures associated with reproduction in the bed bug was made by Ribaga (1897) who noted the notch in the apparent fourth sternite of the female and suggested that it and the accompanying spines were concerned with sound production for sex attraction. The following year Berlese (1898a) gave an account of an organ

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internal to the cuticular structure described by Ribaga. This organ has since come to be known either as the organ of Berlese or the organ of Ribaga. Berlese observed that spermatozoa occurred abundantly in this organ and since he believed that the semen was introduced into the genital tract of the female during copulation in a manner typical of other insects, he suggested that the excess spermatozoa migrated from the so-called spermathecae to the organ which he had discovered. Here he suggested they were digested and that they thus provided an additional source of nutrition for the female. This theory of the utilization of excess spermatozoa for nutrition was termed *hypergamesis*. In support of this theory Berlese pointed out that the ovary does not develop fully until after insemination. In a second paper Berlese (1898b) described digestion of the spermatozoa by the cells of the oviducts in addition to those of the special organ which he had described previously.

At the suggestion of Berlese, Carazzi (1902) undertook a study of the fate of the spermatozoa in the organ of Berlese. As a result of this study, Carazzi seriously questioned Berlese's observation of the digestion of spermatozoa and suggested that Berlese was too much influenced by his theory of hypergamesis. However, Berlese (1909) later included in his book, *Gli Insetti*, a reaffirmation of his views on hypergamesis and lightly passed over the criticisms of Carazzi. Subsequently, various workers studying the reproduction of the bed bug have devoted a very considerable effort either in support or refutation of this theory, and even today the question does not appear to be entirely settled.

Patton and Cragg (1913) described for the first time the actual mode of copulation of *C. lectularius*, showing that the semen is introduced into the organ of Berlese rather than into the genital tract, as Berlese had supposed. Cragg (1914) further described copulation and traced the migration of the spermatozoa from the organ of Berlese, through the hemocoel and eventually to the ovaries.

Murray (1914) published a paper on the anatomy of the bed bug in which he expressed doubt about Cragg's observations on the manner in which copulation occurred. However, Murray had not himself observed copulation and his paper contributed very little to the knowledge of the reproductive organs. Hase (1918) published work on the copulation of the bed bug confirming many of Cragg's observations.

Cragg (1920) published a more detailed study of the migration of the spermatozoa in the female and described some of the histology of the female reproductive organs. He noted that the cells of the organ of Berlese undergo a cyclic change as if a secretion were formed. He also suggested that the spermatozoa pass through the body wall into the organ of Berlese by way of hollow cuticular spines.

This theory of the manner in which the spermatozoa pass through the body wall was further supported in a paper published two years later by Christophers and Cragg (1922). However, the principal object of their investigation was the determination of the homologies and mode of development of the genitalia of both sexes. In this work it was shown that the claw-like appendage of the male designated by Landois (1868) and subsequent investigators as the penis, was actually the left clasper (gonostylus) and that this appendage served as a guiding sheath for the true intromittent organ. In addition, the proper numbering of the abdominal segments and designation of the female gonopods was established in this study.

The egg-laying habits of the bed bug have been the subject of study of several workers. One of the earlier investigations was made by Hase (1919) who noted the effect of various environmental factors on egg production and noted conditions leading to the production of sterile eggs. An investigation of this type was also made by Cragg (1923) but with particular attention given to the influence of the frequency of mating and nutritional state of the male on the number of eggs laid. In this study Cragg believed that he found some evidence in support of the theory of hypergamesis. However, later papers by Titschack (1930) and Johnson (1941) show that Cragg worked with too few individuals to reach valid conclusions.

Titschack (1930) studied the feeding habits of the bed bug in relation to growth and egg production. He demonstrated that the number of eggs produced per milligram of blood consumed depends on various factors such as the age of the female and weight of each egg.

In a very extensive ecological study of the bed bug Johnson (1941) included a very thorough consideration of egg-laying under conditions approximating the natural environment. In the case of the mated, then sexually isolated female, he has shown that there is a gradual decline in egg production with a corresponding percentage increase in sterile eggs until several weeks later all egg production stops. When the female is mated again normal egg production is resumed and the percentage of sterile eggs becomes rather small and remains constant.

Many of the observations made by Johnson have recently been confirmed by Khalifa (1952). In addition Khalifa has shown that unlike its effect on *Rhodnius*, starvation of the male bed bug has little effect on fertility.

The impressively large quantity of spermatozoa received by the female and the ultimate fate of the bulk of it had greatly interested first Berlese, then Cragg. This problem was again taken up by Abraham (1934) who in addition to confirming many of the earlier observations on the course of migration of the spermatozoa, concerned himself with the problem of sperm activation, attraction,

and especially with the sites at which he believed the excess spermatozoa are resorbed. He concluded that the so-called spermathecae function primarily in sperm resorption. This conclusion is questioned by Mellanby (1939) who believes these organs function in sperm storage.

In his study of fertilization and egg production in the bed bug, Mellanby found evidence indicating that egg production in the fertilized female is controlled by the taking of a meal. He also found evidence that indicated that ovarian development is dependent on the presence of spermatozoa in the female rather than on the release of a substance by the organ of Berlese. He also reported that the sperm remain viable in the female for a definite period and that this period is independent of the number of eggs laid. He found no evidence in support of the theory of hypergamesis.

The existence of the organ of Berlese in other members of the family Cimicidae has been studied by Jordan (1922) who found it present in various genera but also found that it varied in position, being either dorsal or ventral and being either median or to the left or right according to the particular species studied. In some Cimicidae no organ of Berlese was found.

Carayon (1946) has demonstrated that the reproductive process of the cimicids *Oeciacus hirundinis* (Jen.), *Leptocimex boueti* (Brumpt), *Ornithocoris toledoi* Pinto, and *Ornithocoris uritui* Lent and Ab. is essentially the same as that in the common bed bug. Carayon (1953c) has also shown that the cimicid *Aphraniola orientalis* Ferris and Usinger has an organ of Berlese although there is no external modification of the body wall indicating its position. Carayon's work suggests that an organ of Berlese may also actually be present in those cimicids in which it has been formerly considered to be lacking.

The family Anthocoridae is generally considered rather closely related to Cimicidae and therefore it has been of interest to know whether or not a mode of reproduction similar to that found in the cimicids occurs among the anthocorids. In the anthocorids studied, Abraham (1934) found no structure corresponding to the organ of Berlese. However, this family has been studied in more detail by Carayon (1952a) who has found that in the subfamily Lyctocorinae there are some species in which the mode of reproduction is essentially the same as in Cimicidae. In other members of this subfamily an organ of Berlese is lacking but the spermatozoa are introduced into the hemocoel by perforation of the genital chamber. In the anthocorid *Xylocoris galactinus* (Fieb.) Carayon (1953a) has found that the organ of Berlese is differentiated into a lobe which receives the sperm and a lobe which conducts the sperm into the body cavity.

In the subfamily Anthocorinae Carayon (1953b) has found a somewhat different means of insemination. In members of this group, the pouch into which the spermatozoa are introduced is joined to the base of each oviduct by a cord of tissue. The spermatozoa do not enter the hemocoel but migrate by way of the tissue bridge to the walls of the oviducts and thence to the ovarioles.

MATERIALS AND METHODS

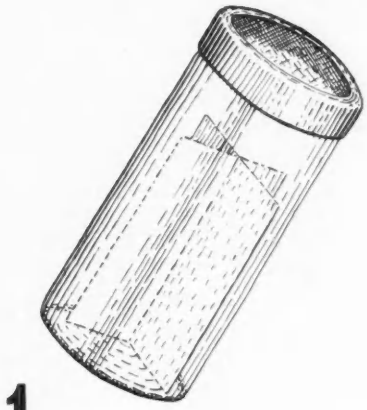
The bed bugs reared in this study were originally obtained from a natural infestation in the animal house of the Department of Zoology of the University of Wisconsin. Several methods of feeding and rearing the insects were tried before a method was finally adopted which seemed to be the most satisfactory for the conditions of this study.

The stock cultures of the bed bugs were kept in wide-mouth specimen jars with a diameter of two inches and a depth of four inches (fig. 1). A piece of heavy blotter paper was placed in each jar to provide a surface on which the bugs could crawl and oviposit. This blotter paper also served to take up the excess moisture. The tops of the metal lids of the jars were removed and the lids then screwed onto the jars over a piece of nylon net. The nylon net was fine enough to prevent youngest nymphs from escaping through it. This type of covering for the culture jars was used so that the insects could feed through the nylon net when the open end of the jar was placed against the host animal.

The blotter paper in the jar did not extend to the top of the jar, preventing the bugs from crawling up onto the nylon cover. This arrangement was found to be convenient since it considerably reduced the number of bugs clinging

LIST OF ABBREVIATIONS

Adg, aedeagus; *AdM*, adductor muscle; *AF*, apical filament; *Bld*, blastoderm; *Ch*, chorion; *Cnx*, connexivum; *CS*, cuticular spines; *DE*, ejaculatory duct; *DPM*, dorsal phallic muscle; *DrM*, dorsorotator muscle; *E*, egg; *EC*, end chamber; *EjP* ejaculatory pump; *EM*, extensor muscle; *EN*, egg nucleus; *EP*, egg plug; *Epd*, epidermis; *Ept*, epithelium; *FB*, fat body; *FE*, follicular epithelium; *FM*, flexor muscle; *Gap*, gonapophyses; *GCh*, genital chamber; *GCM*, genital chamber muscle; *Gcp*, gonocoxopodite; *Grm*, germarium; *Gs*, gonostylus; *HcC*, hemocytoid cell; *Mcl*, muscularis; *Mct*, mycetome; *Md*, mesedenes; *MdR*, mesedenial reservoir; *NtC*, nutritive cord; *OB*, organ of Berlese; *Oc*, oocyte; *OdC*, common oviduct; *OdL*, lateral oviduct; *Ov*, ovary; *Ovl*, ovariole; *Ovp*, ovipositor; *Pd*, pedicel; *PE*, peritoneal epithelium; *PfT*, prefollicular tissue, *Plb*, phallobase; *PS*, peritoneal sheath; *Ra*, ramus; *Rec*, rectum; *SL*, suspensory ligament; *SmN*, segmental nerve; *Sp*, spiracle; *SpC*, sperm cyst; *SpM*, sperm mass; *Spr*, spermatozoa, *SpT*, sperm tract cell; *SR*, sperm reservoir; *Stp*, stapes; *TpB*, tergopleural boundary; *TpM*, tergopleural muscle; *Tr*, trachea; *Ts*, testes; *TsT*, testicular tubule; *Vc*, vacuole; *VD*, vas deferens; *VE*, vas efferens; *VLM*, ventral longitudinal muscle; *VPM*, ventral phallic muscle; *VrM*, ventrorotator muscle; *Vs*, seminal vesicle; *Vtl*, vitellarium; *Vk*, yolk.



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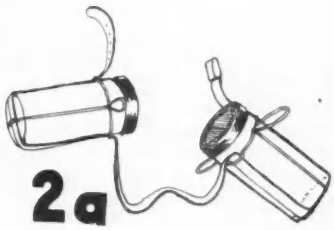

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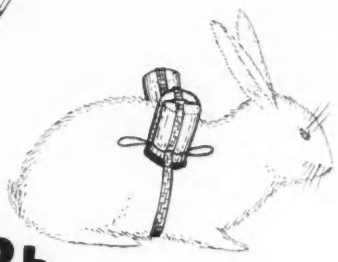

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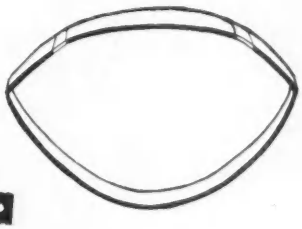

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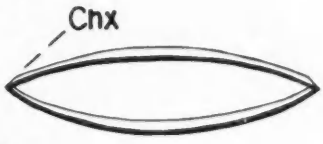

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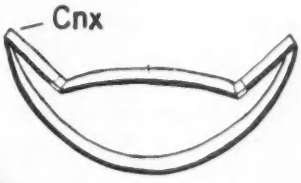

4b

FIG. 1. A culture jar used for rearing bed bugs.

FIG. 2a, b. Culture jars and holder used for feeding bed bugs on a rabbit.

FIG. 3a, b. Diagrammatic cross-sections of the abdominal wall of the bed bug showing the shape when the gut is full and when it is empty.

FIG. 4a, b. Diagrammatic cross-sections of the abdominal wall of a typical heteropteran showing the shape when the gut is full and when it is empty.

(See page 468 for explanation of abbreviations.)

to the lid, making it easier to open the jars without insects escaping. When the jar was opened, the few bugs on the lid were knocked back into the jar and since the bed bug cannot crawl up a vertical glass surface, the lid could be left off without risk of the bugs escaping. On the other hand, when the jar was inverted for feeding the blotter slid down against the cover enabling the bugs to cling to it while feeding through the nylon net.

A special holder for the jars was prepared that made it possible to strap a jar on to each side of a rabbit. This holder consisted of a belt running through wire frames into which the jars fitted (fig. 2a, b). Arms extending anteriorly and posteriorly from the base of the wire frames prevented the jars from being tipped over. The fur of the rabbit was clipped away from the area where the culture jars pressed against the sides so that the bed bugs could more easily penetrate the skin to get their blood meal.

By using this type of feeding arrangement it was not necessary to immobilize the rabbit and thereby the discomfort was minimized. The rabbit gave no indication of experiencing any pain while the bugs were feeding. No damage was done to the area of skin where the bugs fed week after week. The only noticeable effect on the skin where the bugs fed was a slight inflammation lasting a short time after feeding.

The insects were fed once a week. Each jar was left strapped to the rabbit for about two hours so that the entire colony had ample time to feed. When not being fed the insects were kept in an incubator at a temperature of about 30° C. The relative humidity was maintained at about 70 percent by placing large pans of a saturated solution of sodium chloride in the bottom of the incubator.

From time to time the exuviae were dumped from the culture jars. Also it was occasionally necessary to eliminate some of the insects when the colonies became too crowded. It was found that in overcrowded conditions the feces are often ejected accidentally onto one insect by another. In such conditions the feces may dry over the anus and since the animal cannot defecate death eventually results.

A large excess of adult males in a colony may also lead to its decimation. Such a situation may be produced when adult females are being used in an investigation and are removed from the colony. The females remaining in the colony soon die, probably as a result of excessive mating. Since the sperm enters the body cavity, an excess of it can be very harmful. As time goes on the excess of males will increase and the colony declines rapidly.

Because of the rather small size of the bed bug especially fine dissection instruments were required. Dissection needles were prepared by inserting *minuten nadeln* into the ends of match sticks. Dumont Fils number 3c watchmaker's forceps were found to be very satisfactory. It was found that if the insects were fixed before dissection, the dissection was made very difficult because of the adhesion of organs and because the blood-filled gut was very easily ruptured. Therefore the abdomen was always first opened up in insect physiological saline and the gut carefully removed. The insect was then either dissected in the physiological saline or dissected after having been fixed in Bouins.

The anatomy as well as the histology was studied by preparing serial frontal, sagittal and cross sections of the abdomen. Prior to fixation the abdomen was opened up in physiological saline and the gut removed. For ordinary histological studies the tissues were fixed in Bouin's fixative, dehydrated by the alcohol and dioxane method, infiltrated with Fisher's Tissuemat paraffin, and stained with iron hemotoxylin. The difficulties of sectioning the cuticle were reduced to a minimum by using Tissuemat with a high melting point (60° to 62° C.).

In certain tissues in which the cytoplasm is very basophilic or in which the nuclei are very compact the iron hematoxylin stain was unsatisfactory. For such tissues the Feulgen reaction was found to be very useful. The reagents used in the Feulgen reaction were prepared according to the method given by Gomori (1952). The tissue was fixed in acetic alcohol (one part glacial acetic acid to three parts absolute alcohol) and the sections or squashes hydrolyzed for 10 minutes at 60° C. They were then treated with Schiff's reagent for 45 minutes and immersed 5 minutes each in three changes of the sulfite rinse. Fast green was occasionally used as a counter stain.

GENERAL MORPHOLOGY OF THE ABDOMEN

Typically the abdomen of the bed bug is more or less flattened dorsoventrally and appears ovate from the dorsal aspect. In general the abdomens of the male and female are similar in shape but that of the male is smaller and more tapered caudally. The form in both sexes is somewhat influenced by the amount of food that is in the intestine. The dorsal and ventral surfaces of the abdomen meet laterally to form angular

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EXPLANATION OF PLATE II

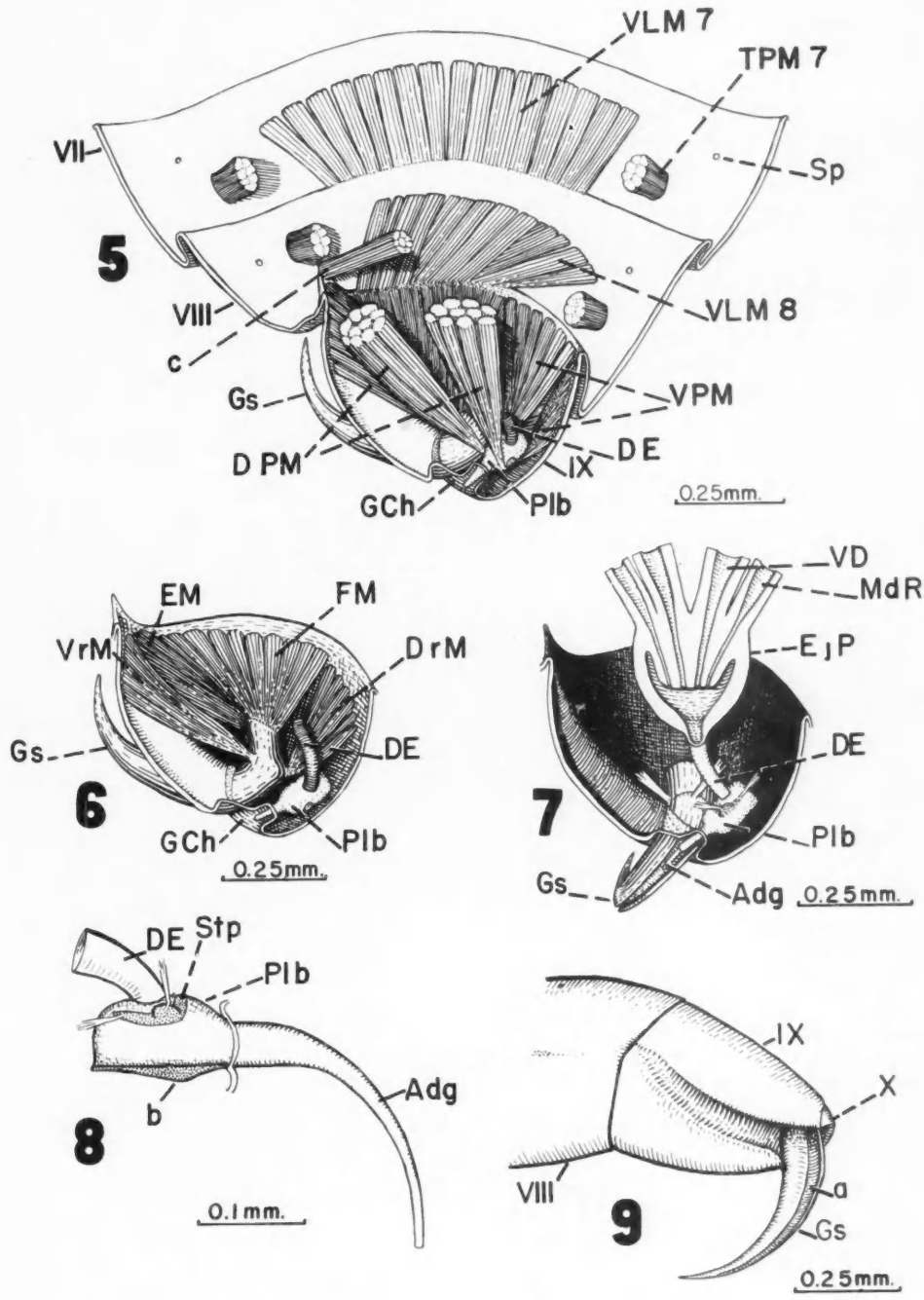
FIG. 5. Dorsal view of the ventral abdominal wall of the terminal segments of the male bed bug showing the musculature.

FIG. 6. Dorsal view of the genital capsule of the male bed bug showing the musculature of the gonostylus.

FIG. 7. Dorsal view of the genital capsule of the male showing the phallus, the extended gonostylus, and optical frontal section of the ejaculatory pump.

FIG. 8. Left lateral view of the phallus with the aedeagus extended.

FIG. 9. Left lateral view of the tip of the abdomen of the male with the gonostylus extended.



(See page 468 for explanation of abbreviations.)

margins, the *connexiva* (fig. 3b, *Cnx*) which typify the heteropteran abdomen. The sclerites of the dorsum of the abdomen extend between the connexiva as continuous sclerotized plates (fig. 3a, b). In contrast these dorsal sclerites of other Heteroptera typically are subdivided by lines of articulation into broad median and small lateral sclerites (fig. 4a, b). This difference is associated with the fact that the mechanism of dorso ventral expansion and contraction of the abdomen of the bed bug is not the same as is often found in Heteroptera. In both the abdomen is more or less elliptical in cross section when the insect is engorged (fig. 3a, b). As the intestinal volume decreases in the typical heteropteran, the median tergal plates move ventrally so that the dorsum becomes concave. In cross section the abdomen is then cresent-shaped (fig. 4b). On the other hand, the dorsum and venter of the abdomen of the bed bug tend to flatten together as the intestine empties. The abdomen of the bed bug is then fusiform when viewed in cross section (fig. 3b). This type of dorsoventral contraction of the abdomen of the bed bug allows the abdomen to flatten much more than that of the typical heteropteran and is probably related to the bed bug's habit of crawling into cracks and crevices for protection.

The spiracles of insects are generally reliable in indicating the approximate boundary between the tergum and pleuron. In the bed bug the spiracles are located on the ventrolateral abdominal surface of the second through eighth segments (fig. 10, *Sp*). Although the spiracles in insects have in some cases been subject to migration, they are generally associated with the lateral margins of the terga. Therefore the definitive margins of the terga of the bed bug abdomen are probably not the connexiva but may be represented by hypothetical lines extending longitudinally through the ventrolateral regions of the abdomen and in the vicinity of the rows of spiracles (fig. 10, *TpB*).

Essentially the same relationship of the tergopleural boundaries to the connexiva and spiracles has previously been discussed by Davis (1955) in a study of the female genitalia of mirids. In many reduviids the tergopleural boundaries apparently are indicated by ventrolateral sutures on the abdomen. On the other hand, in certain Heteroptera such as *Oncopeltus fasciatus* (Dallas) the problem of the location of the tergopleural boundaries is complicated by the fact that the spiracles are located dorsally, mesad of the connexiva.

Additional evidence indicating that the tergopleural boundaries are ventral to the connexiva may be seen in the case of the female genital segments of many Heteroptera. In these segments the individual identity of the primitive limb bases of these segments is often retained. Therefore, the tergopleural boundaries can be distinguished and are found to be ventral to the connexiva. This feature can be seen in the eighth segment of the female bed bug which will be described below. It may also be seen in the ninth segment of female mirids (Davis, 1955). In male heteropterans the boundaries of the primitive limb bases are less clear and therefore of little value to the solution of this question.

In *C. lectularius* the sternites and pleurites of the pregenital segments are presumably fused and there is no indication of a boundary between them. Furthermore, each ventral abdominal sclerite is continuous through the spiracular region to the connexivum. Therefore the ventral wall of the abdomen is a composite of at least sternites and pleurites and possibly lateral portions of the terga, or *paratergites*. For practical purposes however the sclerites of the ventral abdominal wall may be designated as *sternopleurites*.

Ventrally a broad, median unsclerotized region extends from the first segment posteriorly into the seventh segment of the bed bug. This unsclerotized area probably is a region formed by a secondary loss of sclerotization and is associated with contraction and distention of the abdomen.

As has been pointed out by Christophers and Cragg (1922) the first abdominal segment of the abdomen of *C. lectularius* is considerably reduced. This reduction appears to be a general characteristic of the abdomen of Heteroptera. Ventrally the first segment of the abdomen is reduced to a membranous region articulating the abdomen to the sternal and pleural regions of the thorax. The tergite of the first segment is sclerotized in the posterior region which is fused to the second abdominal tergite. The boundary between the first and second tergites is indicated by a distinct sulcus. Passing anteriorly the tergite of the first segment gradually becomes unsclerotized, forming an articulation between the abdomen and the tergal region of the metathorax.

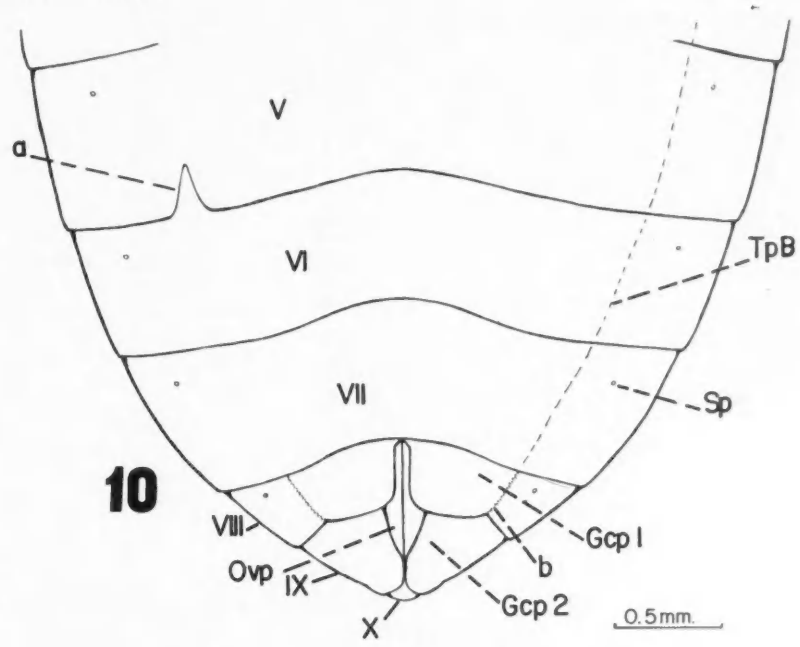
The first through eighth segments of the abdomen of male insects are commonly designated as the pregenital segments. In *C. lectularius* the eighth segment of the male is somewhat modified to accommodate the ninth, or genital segment (fig. 5, *VIII*). The ninth segment of the abdomen must bend sharply to the left during copulation. To enable this bending, the left side of the eighth segment is narrower than the right side.

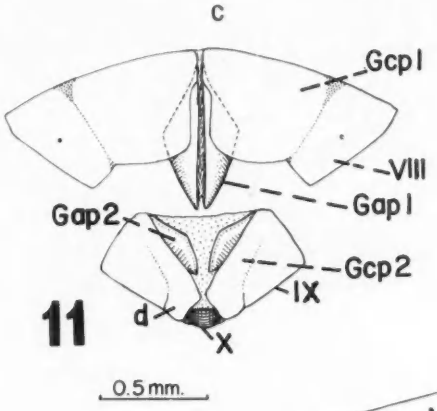
EXPLANATION OF PLATE III

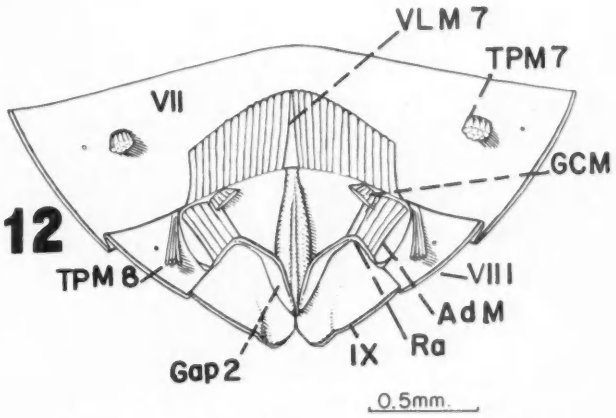
FIG. 10. Ventral view of the posterior ventral abdominal wall of the female bed bug.

FIG. 11. Ventral view of genital segments and genitalia of the female.

FIG. 12. Dorsal view of the ventral wall of the terminal segments of the female showing the genital musculature.







(See page 468 for explanation of abbreviations).

Consequently the tip of the abdomen is somewhat asymmetrical.

The ninth segment forms the cup-like tip of the abdomen. It will be described more fully in the section devoted to the male genitalia. Set into the posterior tip of the ninth segment and slightly dorsad and to the left is the small, narrow, arch-like sclerite of the tenth segment (fig. 9, *X*). This segment forms the roof over a slight invagination in which the anus lies. The eleventh segment is not clearly indicated but may be represented by small arcuate sclerites forming the dorsal and ventral lips of the anus.

The first through seventh abdominal segments of female insects are commonly referred to as the pregenital segments. The fifth and sixth segments of the female bed bug are not in the strictest sense pregenital since the organ of Berlese is associated with these segments. The posterior margin of the fifth segment is notched on the right side beneath the organ of Berlese (fig. 19, *a*). The modifications of the body wall associated with the organ of Berlese will be considered in more detail in the section devoted to this organ.

In many heteropterans and other insects the posterior margin of the seventh segment of the female projects medially over the base of the ovipositor forming what is usually designated as the *subgenital plate*. This structure is not present in *C. lectularius*.

The eighth and ninth segments of the abdomen of the female, the genital segments, and their appendages will be discussed more fully in the section dealing with the female genitalia. The postgenital segments are greatly reduced in *C. lectularius*. The tenth segment is a narrow, sclerotized arch beneath the posterior margin of the tergite of the ninth segment and forms a roof over the anus (fig. 14, *X*). Mid-ventrally this segment is not sclerotized and laterally it is not differentiated into connexiva. The presence of the eleventh segment is not clearly indicated in the abdomen of the female of *C. lectularius*. As in the male it is possibly represented by small arcuate sclerites forming the dorsal and ventral lips of the anus (fig. 14, *XI*).

Morphology of the Genitalia

The general concepts of the origin and evolution of the genitalia of insects which serve as a basis for the morphological considerations in this study are, for the most part, based on the extensive studies of Snodgrass (1933, 1935, 1936), Michener (1944), and Gustafson (1950). Although the views of these authors differ in several major respects, the differences do not have a serious bearing on the present study.

The terminology used in designating the structures of the male genitalia is essentially the same as used by Bonhag and Wick (1953) in their study of *Oncopeltus fasciatus*. The terminology of the female genitalia is essentially the same as used by Michener (1944) and Gustafson (1950).

GENITALIA OF THE MALE

In Heteroptera the ninth segment of the male is characteristically in the form of a bulbous sclerite which is termed the *genital capsule*. This segment bears the genital claspers and the intromittent organ. The postgenital segments form a short tubular *proctiger* which lies dorsally over the genital capsule. The genital claspers are well developed in most Heteroptera. Bonhag and Wick (1953) have concluded that these claspers are modified styli and therefore terms them *gonostyli*. This designation is used in this paper although it should be noted that the evidence indicating that the claspers are styli rather that *parameres* does not appear to be entirely conclusive.

The intromittent organ, or *phallus*, of the Heteroptera is generally differentiated into a basal supporting sheath, the *phallobase* and an extendible tube, the *aedeagus*.

In general the male genitalia of the bed bug are similar to those of other heteropterans. The ninth segment is in the form of a blunt cap at the tip of the abdomen. Its tip is directed slightly to the left. Laterally it is not differentiated into connexiva and the boundaries of the tergal, pleural and sternal regions are not distinguishable. The right gonostylus has been lost (Christophers and Cragg, 1923). The left gonostylus is in the form of a claw-like appendage arising from the tip of the segment (figs. 5, 9, *GS*). At rest its apex is directed anteriorly and the appendage lies in a deep groove (fig. 9) which extends along the left lateral margin of the ninth segment. A slight groove can also be distinguished along the right lateral margin of this segment.

A channel extends from the base of the gonostylus almost to its tip (fig. 9, *a*). Proximally this goove is ventral and passes to the outer margin of the gonostylus.

Dorsal to the base of the gonostylus the tenth segment can be distingusished. This segment forms a complete sclerotized annulus which encloses a small chamber containing the anus (fig. 9, *X*). Between the base of the gonostylus and the tenth segment there is a narrow inconspicuous atrium which is the genital chamber (fig. 6, *GCh*).

When the gonostylus is extended for copulation it moves slightly caudad and is rotated ventrad. In this position its axis almost forms a right angle to the axis of the ninth segment (fig. 9). The

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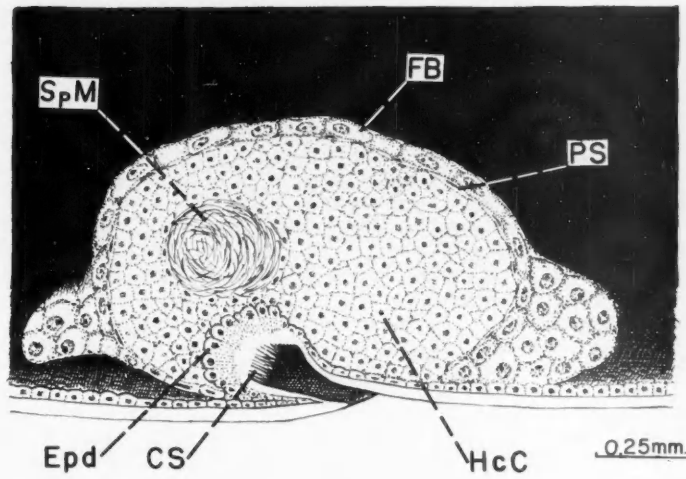
EXPLANATION OF PLATE IV

Fig. 13. Lateral view of an optical longitudinal section through the organ of Berlese.

Fig. 14. Lateral view of an optical sagittal section through the abdomen of the female showing the reproductive organs.

Fig. 15. Semidiagrammatic view of a portion of a lateral oviduct containing an egg and showing the layers of tissue composing the wall of the lateral oviduct.



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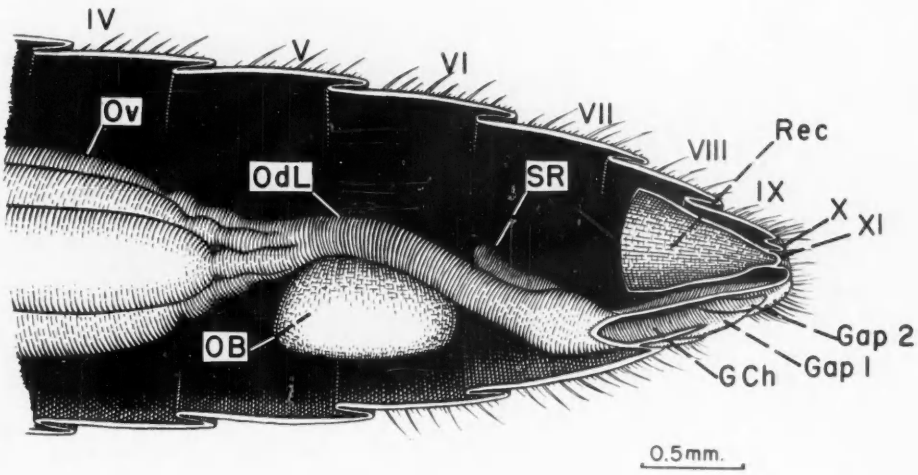

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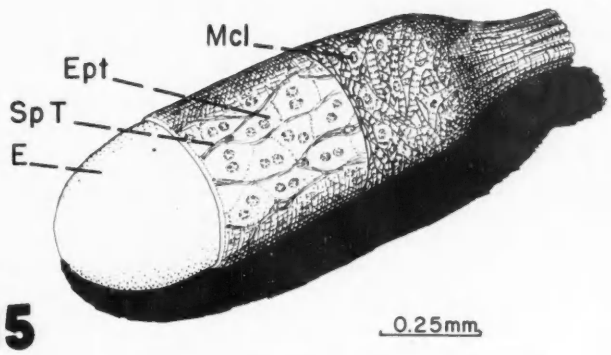

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(See page 468 for explanation of abbreviations.)

base of the channel extending along the gonostylus lies against the phallus when the gonostylus is extended (fig. 7, *Plb*). Thus when the aedeagus is everted it enters the channel and is guided by it to the tip of the gonostylus (fig. 7 *Adg*).

The phallus of the bed bug consists of a basal capsule-like portion referred to here as the *phallobase* (fig. 8, *Plb*) and an extensible terminal portion designated as the *aedeagus* (fig. 8, *Adg*). The phallobase is supported by a sclerotized strip which lies along its left ventrolateral surface and which is joined to the posterior margin of the genital capsule (fig. 8, b). On the roof of the capsule-like phallobase there is a U-shaped sclerite the arms of which extend anteriorly (fig. 8, *Stp*). This sclerite is designated as the *stapes* and is typically a part of the phallobase of Heteroptera. Two pairs of tendons arise from this sclerite and the phallic muscles are inserted on these (fig. 8). The posterior pair of tendons arise from apodemes on the sclerite.

The aedeagus, is a long narrow tubular structure whose walls are not sclerotized (fig. 8, *Adg*). When not in use the aedeagus is telescoped into the capsule-like phallobase. It is probably extended by fluid pressure.

THE GENITAL MUSCLES OF THE MALE

The knowledge of the musculature of the genitalia of Heteroptera is not sufficiently well established to enable the determination of homologies. Therefore the designations of the genital muscles given below are purely descriptive and have no special morphological significance.

The principle muscles of the pregenital segments are the dorsal and ventral longitudinal muscles (fig. 5, *VLM 7*) and a pair of tergopleural muscles extending dorsoventrally between the tergite and sternopleurite of each segment (fig. 5, *TPM 7*).

The ventral longitudinal muscles of the eighth segment are especially well developed (fig. 5, *VLM 8*). Their insertion on the ninth segment is displaced to the left so that they lie diagonal to the axis of the body. The dorsal longitudinal muscles of the eighth segment are less well developed but are also diagonally arranged.

A muscle apparently related to the dorsal longitudinal muscles of the eighth segment originates medially on the eighth tergite and extends diagonally to its insertion on an apodeme projecting from the left anterior-lateral margin of the ninth segment (fig. 5, *c*.)

The action of these muscles inserted on the ninth segment, or genital capsule, is to direct its tip ventrally and to the left in the act of copulation.

In the genital capsule two pairs of muscles are inserted on the phallobase. One pair has its origin anteriorly and dorsally on the genital capsule; these muscles extend diagonally to the right to their insertion on the apodemes of the stapes of the phallobase (fig. 5, *DPM*). These are designated here as the *dorsal phallic muscles*. The second pair of phallic muscles has its origin on the right ventrolateral region of the genital capsule; they are inserted on the anterior tendons extending from the stapes (fig. 5, *VPM*). These are designated as the *ventral phallic muscles*.

The two pairs of phallic muscles serve to direct the phallus so that when the aedeagus is extended it will enter the channel of the left gonostylus. They probably also rotate the phallus to coincide with the rotation of the gonostylus as it is extended.

Four muscles are inserted on the gonostylus. On the right a muscle originates on the anterior lateral region of the genital capsule and is inserted on a tendon arising from the dorsal surface of the apodeme of the gonostylus (fig. 6, *VrM*). This is designated here as the *ventrorotator muscle*. Immediately anterior to this there lies another muscle which originates ventrolaterally on the genital capsule and which is inserted on the apex of the apodeme of the gonostylus (fig. 6, *EM*). This muscle is designated here as the *gonostylus extensor muscle*. The third muscle of the gonostylus is median and originates on the anterior ventral region of the genital capsule (fig. 6, *FM*). It is inserted on the base of the gonostylus apodeme. This muscle is designated here as the *gonostylus flexor muscle*. On the right ventrolateral aspect of the genital capsule a small fan-shaped muscle has its origin. It is inserted laterally on the apodeme of the gonostylus (fig. 6, *DrM*). This muscle is designated here as the *dorsorotator muscle* of the gonostylus.

The ventrorotator and extensor muscles move the gonostylus posteriorly and ventrally into the erect position. The dorsorotator and flexor muscles return the gonostylus to its resting position in the groove along the left side of the genital capsule. The antagonistic action of the dorso- and ventrorotators and of the flexor and extensor muscles serves to hold the gonostylus in a rigid position when erect.

GENITALIA OF THE FEMALE

In most families of Heteroptera all of the basic parts of the ovipositor may be distinguished although in many families these parts may be greatly reduced. The supporting base of the ovipositor consists of a pair of sclerites, the *first* and *second gonocoxopodites* (valvifers) on the venter of eighth and ninth segments respectively. These sclerites are generally considered to be the remnants of the limb bases of the abdominal appendages of primitive insects. The blades of the ovipositor are a pair of processes extending from each pair of gonocoxopodites and are termed the *first* and *second gonapophyses* (valvulae). Another pair of processes, the *third*

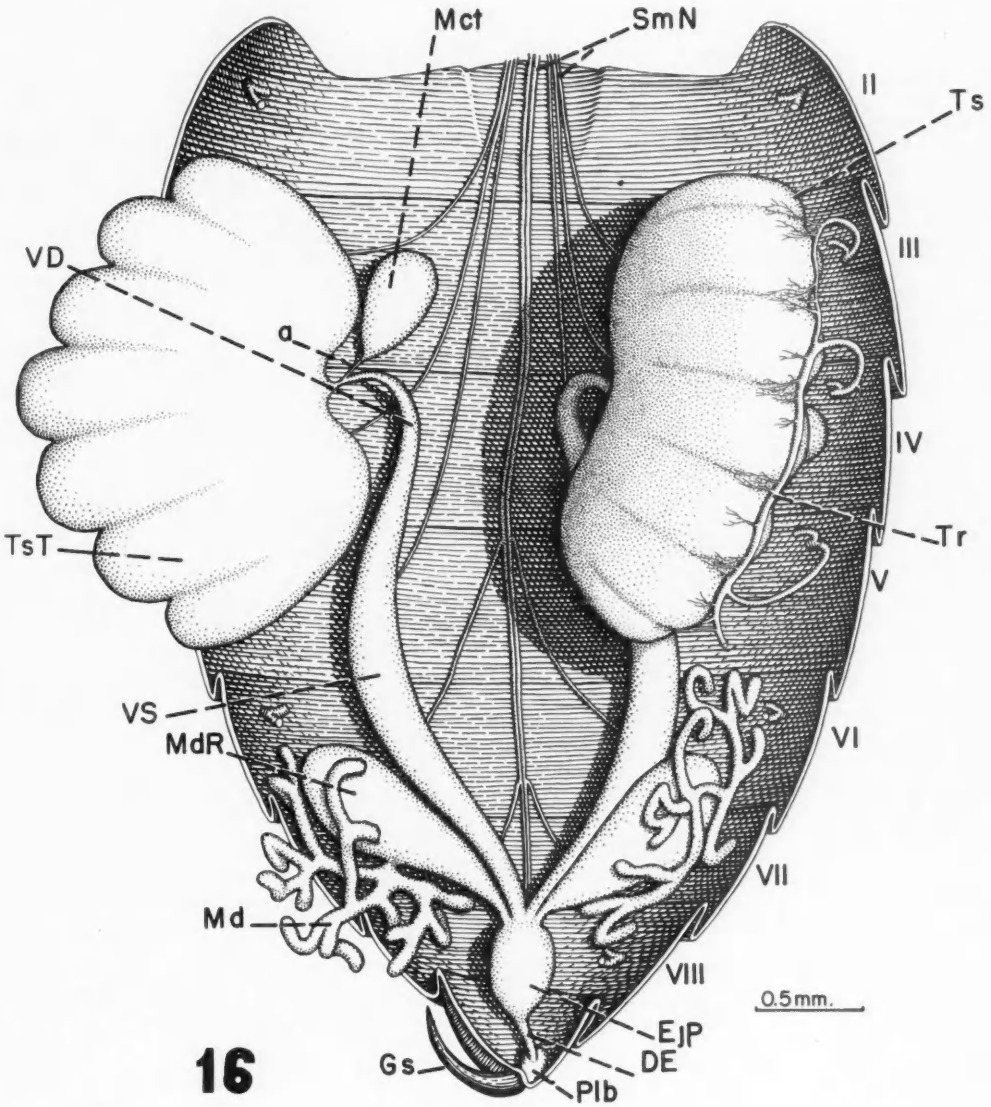

FIG. 16. Dorsal view of the ventral abdominal wall and male reproductive organs with the left testis laid to one side to expose the mycetome.

(See page 468 for explanation of abbreviations.)

gonapophyses, also extend from the second gonocoxopodites in many insects. These are often distinguishable in heteropteran females but never serve as blades of the ovipositor.

The first gonocoxopodites of the bed bug are more or less quadrate sclerites broadly joined to the inflected tergum of the eighth abdominal segment (fig. 11, *Gcp 1*). These sclerites have been referred to as *admedian plates* by Rothschild (1912) and were considered to be sternites by Christophers and Cragg (1922). The suture between each first gonocoxopodite and the inflected tergum represents the tergopleural boundary (fig. 10, *b*) and is probably equivalent to a part of the ventral connexival suture found in many other heteropterans.

At the anterior mesal angles the first gonocoxopodites are joined by a narrow strip of membranous cuticle which may possibly represent what is left of the very reduced eighth sternite (fig. 11, *c*).

The gonapophyses of the first gonocoxopodites are rather short, cone-shaped processes (figs. 11, 14, *Gap 1*). Proximally each gonapophyses tapers into a thin ramus which joins the gonapophysis to the mesal anterior angle of its gonocoxopodite. The first gonapophyses are joined together by a strip of membranous cuticle which is a continuation of the membraneous sternal region between the first gonocoxopodites.

In the ninth abdominal segment the second gonocoxopodites are fused laterally to the tergites and their mesal margins overlap the ovipositor (fig. 11, *Gcp 2*). The very reduced second gonapophyses of these gonocoxopodites are small flat projections (fig. 11, *Gap 2*) and each is joined to the anterior margin of its gonocoxopodite by a short, thin ramus (fig. 12, *Ra*). The second gonapophyses lie dorsal to the posterior portion of the first gonapophyses (fig. 14, *Gap 1*, *2*) and are not locked with the first by a sliding tongue-and-groove mechanism as is commonly the case in more specialized ovipositors.

The third gonapophyses are reduced beyond clear recognition. They are probably represented by protuberances at the posterior end of each of the second gonocoxopodites (fig. 11, *d*).

THE GENITAL MUSCLES OF THE FEMALE

The reduced condition of its ovipositor is probably related to the egg-laying habits of the bed bug. The eggs are deposited on and gluded to surfaces rather than being buried. As one might expect, the movements of such an ovipositor are rather simple and the musculature is correspondingly reduced. The principle set of muscles concerned with movements of the gonapophyses originate near the lateral anterior margin of each of the first gonocoxopodites and are inserted on the anterior mesal angle of each of the second gonocoxopodites (fig. 12, *AdM*). This pair of muscles will be designated as the *adductor muscles* of the second gonapophyses. Mesad of the adductor muscles another pair originate on the first gonocoxopodites (fig. 17, *GCM*). These muscles fan out from their origin to insert on the dorsal lateral surfaces of the genital chamber. This pair of muscles is designated as the *genital chamber muscles*.

Two other pairs of muscles are associated with the first gonocoxopodites. The ventral longitudinal muscles originating on the seventh segment are inserted on the anterior edge of the first gonocoxopodites (fig. 12, *VLM 7*). The tergopleural muscles of the eighth segment are attached to the lateral anterior angle of each of the first gonocoxopodites (fig. 12, *TPM 8*). No tergopleural muscles were found in the ninth segment.

In the process of oviposition the egg is first passed into the genital chamber by the wave-like contraction of the muscularis of the oviducts. As the egg enters into the genital chamber it is forced down between the blades of the ovipositor by the paired genital chamber muscles. The egg is then passed through the ovipositor by the movements of the second gonapophyses. These movements are brought about by their adductor muscles.

The musculature of the ovipositor of heteropterans is not sufficiently well known to establish homologies. Judging from those studies describing this musculature, it appears that the genital musculature of the female bed bug is very dissimilar to that of many other heteropterans.

THE ORGAN OF BERLESE

As noted earlier, Ribaga (1897) and Berlese (1898) did not clearly understand the function of the organ of Berlese. Patton and Cragg (1913) were first to point out that the organ actually functions in copulation. In a later study of the organ Cragg (1920) noted the roles of Ribaga and Berlese in its discovery. He felt at the time too little was known to name the organ on the basis of its homologies. He therefore referred to the cuticular portion as the *organ of Ribaga* and to the cellular portion as the *organ of Berlese*. Subsequent workers did not make this distinction but referred to the organ either as the organ of Berlese or as the organ of Ribaga. In the present paper the author has chosen to designate it as the organ of Berlese primarily in recognition of the great contribution which Berlese has made to the science of entomology and because Berlese came nearer to realizing the true function of the organ. However the author also feels that if a name based on function had been applied by Cragg, much confusion could have been avoided.

ANATOMY AND HISTOLOGY

As noted previously the position of the organ of Berlese is indicated externally by a deep notch in the right posterior margin of the fifth sternopleurite (fig. 10, *a*). In the conjunctiva at

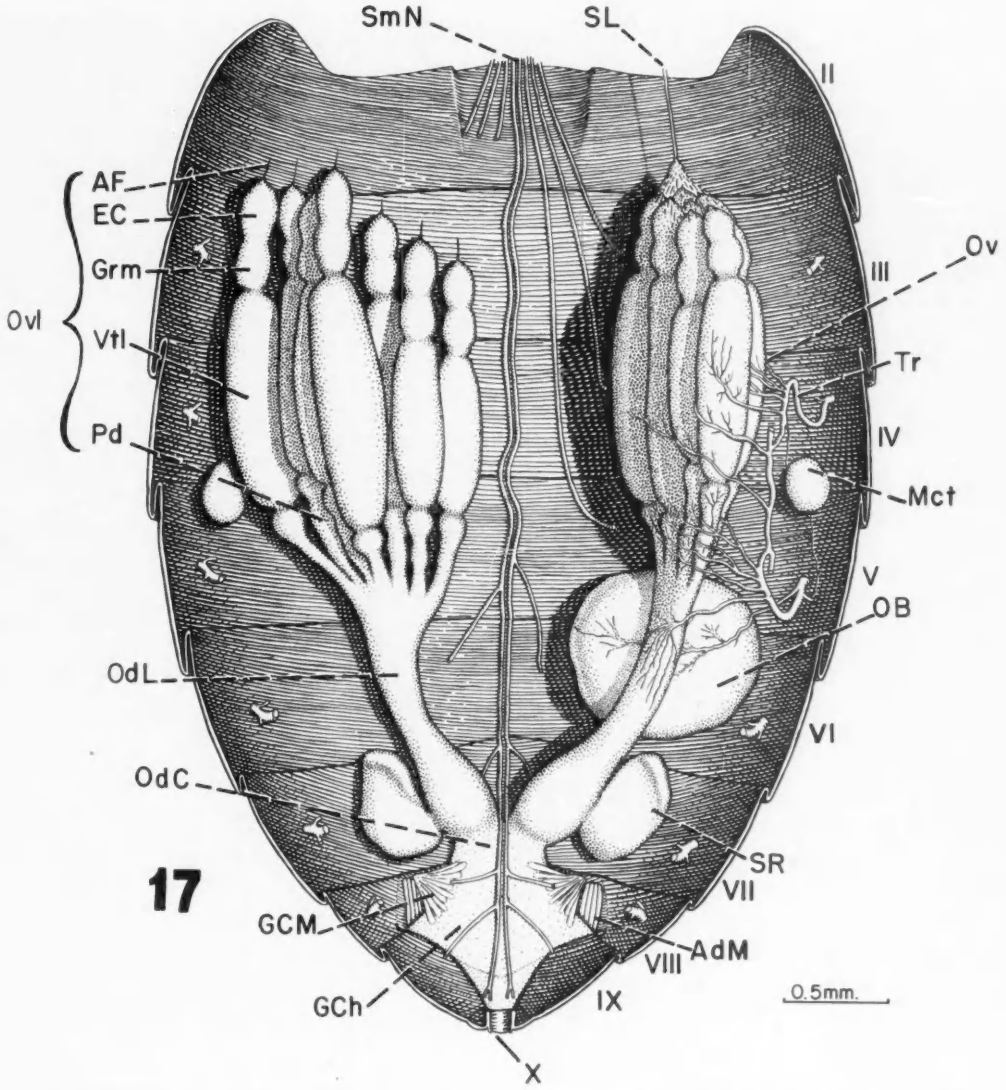

FIG. 17. Dorsal view of the ventral abdominal wall and reproductive organs of the female showing the right ovary in a normal position and the left ovary dissected out to expose the ovarioles.

(See page 468 for explanation of abbreviations.)

the base of this notch there is a shallow cuticular pocket which is surrounded internally by a large mass of cells resembling hemocytes (fig. 13, *HcC*). This mass of cells is encapsulated by a thin peritoneal sheath which is in turn covered by a thin layer of fat body tissue (fig. 13, *PS*, *FB*).

The bottom of the cuticular pocket of the organ of Berlese is lined with a dense covering of minute cuticular spines (fig. 13, *CS*). The cuticle forming the pocket is considerably thicker than the surrounding cuticle of the conjunctiva and consists primarily of a thick endocuticular layer. There is no sclerotization.

The epidermis adjacent to the part of the cuticle lined with cuticular hairs is characteristically modified. The cells, rather than being cuboidal as are typical epidermal cells, are globose and lie at various levels. The apices of the cells extend toward the cuticle and the cells gradually merge with the cuticle so that there is not a distinct boundary between the two (fig. 13, *Epd*).

The cellular mass internal to the cuticular pocket is very loosely arranged so that the shape of the organ is influenced by the fullness of the gut. When fully engorged the gut presses against the Organ of Berlese and flattens it out considerably. When the organ is not pressed upon, it assumes a hemispherical shape.

The organ does not have a lumen or internal cavity but is completely filled with a mass of cells which resemble blood cells and which are therefore designated here as *hemocytoid cells* (fig. 13, *HcC*). The appearance of these cells varies according to the reproductive state of the female. In the virgin female the cells are more or less ovate and there are many interstices. The cytoplasm shows little or no basophilia and the nuceli are relatively small and are spherical.

During the postembryonic development of the organ of Berlese numerous mitotic figures of the hemocytoid cells may be found. A count of the chromosomes of these cells indicates that the cells are diploid at this time. The nuclei do not increase in size during or after postembryonic development of the organ so it is doubtful that endomitosis occurs, and the nuclei of this tissue are probably not polyploid as are the nuclei of many other insect tissues.

Mellanby (1939) has transplanted the organ of Berlese of recently mated females into virgin females and found that there was no ovarian stimulation, an indication that egg production is not initiated by an internal secretion of the organ of Berlese.

INSEMINATION

The cuticular pocket of the organ of Berlese is punctured by the gonostylus of the male so that semen may be introduced. This fact may be demonstrated by dissected pairs anesthetized while mating.

Cragg (1920) and Christophers and Cragg (1922) did not realize that the semen was introduced in this manner but believed that it was deposited into the cuticular pouch. Furthermore they believed that the spines lining the pouch were hollow and that the spermatozoa migrated through the lumen of these spines into the organ of Berlese. They noted that the spermatozoa could be found in a large mass in the organ immediately after copulation and therefore concluded that the migration through the cuticle was indeed very rapid and remarkable. It was apparently realized by Abraham (1934) and clearly recognized by Carayon (1952a) that the cuticle is punctured during copulation. There is now no reason to believe that the spines of the organ of Berlese are hollow and according to Carayon (1953a) they are usually not present in the organ of Berlese of other Cimicidae.

PASSAGE OF THE SPERMATOZOA THROUGH THE ORGAN OF BERLESE

The spermatozoa are injected into the center of the organ of Berlese in a very dense mass which displaces the hemocytoid cells (fig.13, *SpM*). Within a few hours this mass has become diffuse and the spermatozoa gradually make their way through the interstices to the periphery of the organ. Here they accumulate, forming a layer between the hemocytoid cells and the connective tissue sheath. In the region where most of the spermatozoa have accumulated the wall of the organ of Berlese ruptures and the mass of spermatozoa flow out, probably carried in part by the escape of fluid which has accumlated in the organ.

It is of interest to note that in the anthocorid *Xylocoris* Carayon (1953a) has found that the organ of Berlese is apparently differentiated into two parts. In one lobe the spermatozoa are received and they pass from here to a lobe where they are discharged into the body cavity.

FUNCTIONAL SIGNIFICANCE OF THE ORGAN OF BERLESE

The knowledge that the gonostylus penetrates the body wall makes it possible to reach certain conclusions regarding the functional and evolutionary significance of the organ of Berlese. Carayon (1952a) has shown that the type of copulation found in bed bugs probably evolved from an insect which introduced the sperm into

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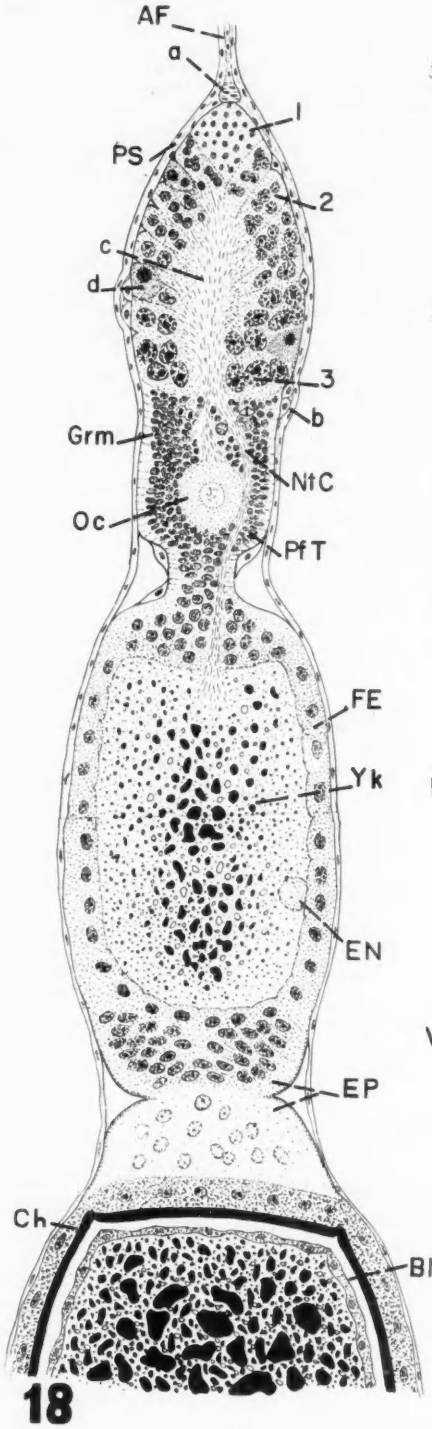
EXPLANATION OF PLATE VII

FIG. 18. A longitudinal section through an ovariole showing the end chamber, germarium, and a portion of the vitellarium.

FIG. 19. A longitudinal section through a testicular tubule of a young male showing the spermatozoa in various stages of development.

FIG. 20. A cross-section through the seminal vesicle.



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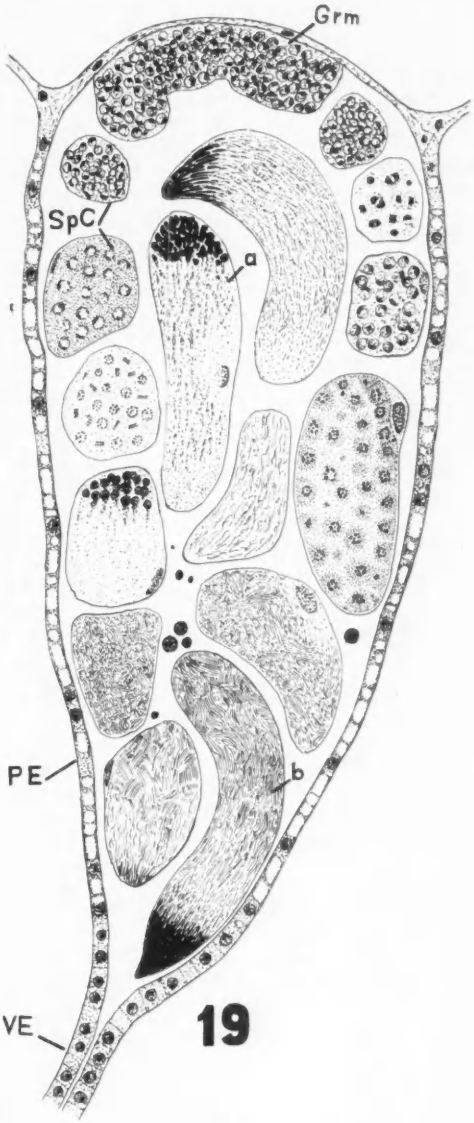

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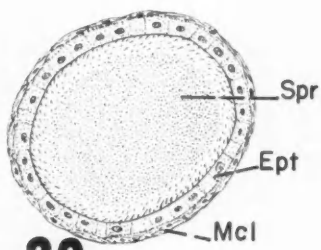

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(See page 468 for explanation of abbreviations.)

the genital chamber to one that punctured the genital chamber and introduced the sperm into the body cavity and finally to one that introduced the sperm into the body cavity through intersegmental membranes of the body wall. It may also be added to this theory that since these organs are found in various locations according to the species concerned, the ancestral forms probably punctured the abdomen of the female at various convenient locations and that eventually various species evolved which may have characteristic specific sites at which this puncture is made.

In *C. lectularius* an organ of Berlese commonly develops on the left side of the abdomen as well as on the right side. The degree to which this vestige develops varies considerably among different individuals. The common occurrence of a rudimentary left organ of Berlese led Cragg (1920) to speculate that these organs were originally the primitive paired gonopores of the female reproductive tract. This seems quite unlikely in view of the knowledge of the evolution of the reproductive organs that has been accumulated since the time that Cragg's suggestion was made.

Christophers and Cragg (1922) have shown that a right gonostylus in the male develops in the fifth instar to a certain point and then disappears. This vestigial gonostylus plus the vestigial organ of Berlese in *C. lectularius* suggests that its ancestor may have been able to mate ambidextrously.

In view of the fact that the body wall is punctured during insemination the organ of Berlese may be logically assumed to have evolved as the result of the hazards involved in making such a break in the cuticle. These hazards would include such things as excessive bleeding at the site of the wound, the introduction of foreign material into the body cavity, and the danger of injury to vital internal organs of the female by the gonostylus.

The structure of the cuticular pocket is such that the puncture may be made with little injury to the female. The arrangement of the epidermal cells is such that they may close over the wound when the gonostylus is withdrawn. The cuticle is soft enough to allow the penetration of the gonostylus but its thickness helps prevent excessive loss of body fluids. The hemocytoid cells probably also aid in the closing of the wound.

The hemocytoid cells probably serve to prevent anything but active spermatozoa from entering the body cavity. Their activity in phagocytosis of the seminal fluid will be described below.

Without an organ of Berlese the danger of the gonostylus injuring internal organs would be particularly critical in an insect such as the bed bug since when it takes a full meal the midgut is pressed tightly against the body wall. The organ of Berlese thus serves as a cushion to protect the midgut from the penetrating gonostylus. It is quite likely that occasionally the cushion is insufficient and that the midgut is punctured nevertheless. Individuals in which the gut has been ruptured are sometimes found in the colonies. These insects become a very deep red all over and die after several days.

PHAGOCYTIC ACTIVITY OF THE ORGAN OF BERLESE

The cyclic changes that the hemocytoid cells undergo after the female mates was first observed by Berlese (1898). However, his observations and interpretations of these changes were influenced too much by his erroneous theory of the function of the organ of Berlese.

A more accurate description of these changes was made by Cragg (1920). In his study it was shown that at the time of copulation many of the cells of the organ of Berlese contain a large vacuole which displaces the nucleus to one side of the cell. At about the time that the spermatozoa leave the organ of Berlese an eosinophilic granular mass appears within the cell vacuoles. This mass becomes more dense and eventually becomes basophilic. After several days the basophilic bodies become smaller and eventually disappear. Cragg concluded from these observed changes that the organ of Berlese is a glandular organ which produces an internal secretion after copulation has occurred. No further studies of the changes in the organ of Berlese have been made and so Cragg's conclusions have not been confirmed.

The activity of the cells of the organ of Berlese described by Cragg have also been observed in

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EXPLANATION OF PLATE VIII

FIG. 21. A section through the tip of a testicular tubule showing a sperm cyst in the process of turning. Iron hematoxylin. Photo approx. 300X.

FIG. 22. A section showing the vacuoles among the hemocytoid cells of the organ of Berlese. Iron hemotoxylin. Photo approx. 300X.

FIG. 23. Whole mount of the lateral oviduct wall showing the sperm tract cells of a recently fed female. Iron hematoxylin. Photo approx. 300X.

FIG. 24. A section through the sperm reservoir of a recently mated female. Feulgen reaction. Photo approx. 150X.

FIG. 25. Whole mount of follicular epithelium. Feulgen reaction. Photo approx. 300X.

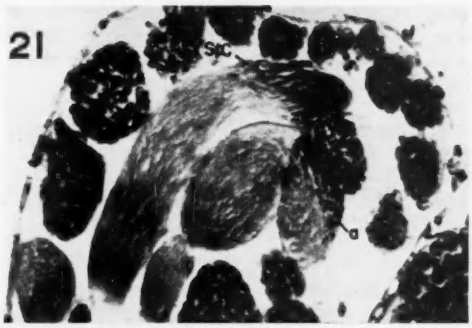
FIG. 26. Longitudinal section through the end chamber and germarium of an ovariole. Iron hematoxylin. Photo approx. 300X.

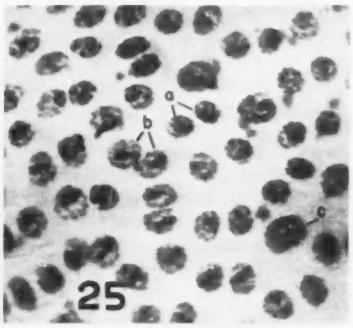
FIG. 27. Same as Fig. 26. Feulgen reaction. Photo approx. 300X.

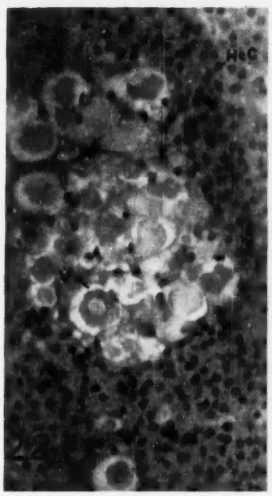
FIG. 28. Whole mount of the lateral oviduct wall showing the sperm tract cells of a starved female. Iron hematoxylin. Photo approx. 300X.

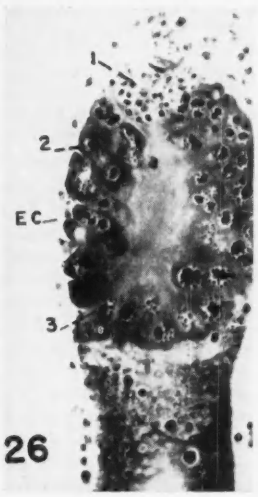
FIG. 29. A section through the sperm reservoir showing a cross-section of a bundle of spermatozoa. Iron hematoxylin. Photo approx. 300X.

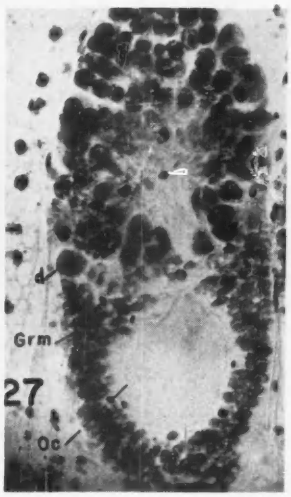
FIG. 30. Longitudinal section of the vitellarium showing the blastoderm and early formation of the chorion. Iron hematoxylin. Photo approx. 300X.

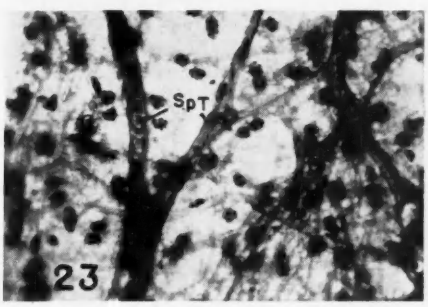


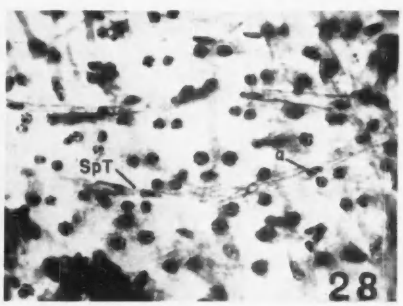


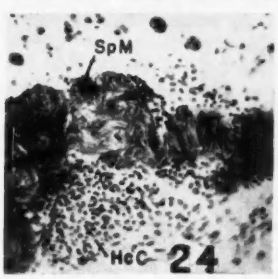


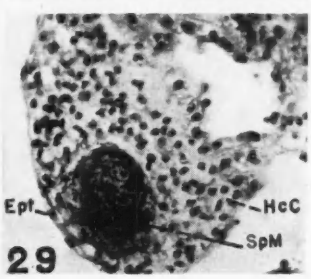


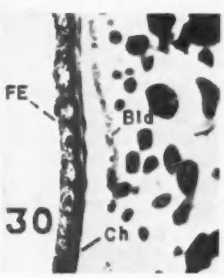












(See page 468 for explanation of abbreviations.)

the present investigation but a different interpretation of their significance has been made. Judging from the histology and staining reactions of the organ of Berlese there is serious reason to doubt that it is secretory in the usual sense of the word.

It has been demonstrated that in other insect tissues the nuclei of secretory tissues are usually highly polyploid (Geitler, 1941; Merriam and Ris, 1954). The nuclei of tissues in the bedbug which are known to be excretory are apparently also polyploid. However, as has been pointed out above, the nuclei of the hemocytoid cells are probably diploid. Furthermore, the cytoplasm of these cells is never basophilic as is commonly true of the cytoplasm of secretory cells.

The activity of the cells of the organ of Berlese following mating may be more properly described as phagocytosis. When the semen is injected into the organ of Berlese a considerable part of the semen consists of the secretion of the male accessory glands. This part of the semen must be disposed of in some manner in the female. It is apparently taken up and digested by the cells of the organ of Berlese (fig. 22, *Vc*). This view is supported by the fact that secretion of the accessory glands has a staining reaction similar to that of the eosinophilic bodies which appear in the cells of the organ of Berlese. Furthermore, if the cells of the organ of Berlese are derived from blood cells as they are reported to be, then a phagocytic activity seems more natural to them than would a secretory activity.

Copulation

When a male and female which have been sexually isolated several days are placed together in the same container and observed in subdued light they may be seen to wander at random until they accidently meet. Sex attraction apparently depends on contact recognition rather than scent, sound, or visual attraction. When they make contact the female usually becomes motionless and lies flat against the surface so that the male must either go around or crawl over her. If the male is sexually active he moves on top of the female, orients himself diagonally and grasps her. The tip of the abdomen of the male is then turned under the right side of the abdomen of the female and the gonostylus is extended and inserted into the cuticular pocket of the organ of Berlese. The female raises the tip of her abdomen slightly thus moving apart the ventral segments and making the entrance to the organ of Berlese more exposed. The female is otherwise absolutely motionless as the male mounts her. When in a suitable position the male pushes the sharp tip of the gonostylus through the cuticular pocket thus puncturing the body wall. The aedeagus, which has passed down the groove in the gonostylus, now opens into the cellular mass of the organ of Berlese wherein the semen is deposited. The pair may remain in copula for one to several minutes. However, after the male has punctured the organ of Berlese, the female no longer remains motionless, but usually moves about actively carrying the male with her.

The Male Reproductive Organs and Their Ducts

General Morphology and Terminology

The two large, fan-shaped testes of the bed bug lie mostly within the third, fourth, and fifth abdominal segments (fig. 16, *Ts*). They are held in position primarily by the many trachea which supply them. As in other Heteroptera each testis consists of seven chambers, the testicular tubules (fig. 16, *TsT*). The product of each testicular tubule passes out by way of a basal efferent duct, the *vas deferens* (fig. 19, *VE*). The vasa efferentia of each testis join a common lateral duct, the vas deferens (fig. 16, *VD*). In the bed bug, as in other heteropterans, a portion of each vas deferens is dilated to form a tubular chamber, the *seminal vesicle* (fig. 16, *VS*) which receives and stores the spermatozoa. Since each vas deferens emerges as a very narrow duct and gradually enlarges, the seminal vesicle is not clearly differentiated from the rest of the duct. In mature bugs the entire duct is compactly filled with spermatozoa which in freshly dissected specimens gives the vas deferens a glistening white appearance.

Posteriorly the vasa deferentia again become narrowed and enter a median bulbous muscular structure, the *ejaculatory pump* (fig. 16, *EjP*). Running posteriorly from the ejaculatory pump is the unpaired *ejaculatory duct* (fig. 16, *DE*) which leads to the intromittent organ.

One or both of two types of male accessory glands may be found in heteropterans. These are the *mesadenia* and *ectadenia*. The mesadenia are mesodermal glands and open into the vasa deferentia. The ectadenia are ectodermal and open into the anterior region of the ejaculatory duct. Both types of glands serve to produce the seminal fluid. In the bed bug only the mesadenia are present (fig. 16, *Md*). Each consists of several branched tubules which in general appearance somewhat resemble the Malpighian tubules. At its base each gland opens laterally into its reservoir (fig. 16, *MdR*). The *mesadenial reservoirs* are thin-walled oblong sacs lying laterally alongside of each seminal vesicle. Posteriorly each reservoir gradually narrows to form a duct which enters the ejaculatory pump laterad of each vas deferens. In fresh preparations the reservoirs may be seen to contain the clear, colorless secretion of the mesadenia.

A small spherical or pyriform organ (fig. 16, *Mct*) is found attached by a strand of connective

tissue (fig. 16, *a*) to the base of each testis. This organ is the ***mycetome*** and contains the rickettsial symbiotes of the bed bug. It was first observed by Patton and Cragg (1913) who designated it as an accessory lobe of the testis. In 1921, Buchner rediscovered this organ and demonstrated that it contained symbiotes. In 1923, Buchner made a thorough study of the histology and development of the mycetome. However the designation given by Patton and Cragg still appears in certain entomology texts, and in the recent study made by Khalifa (1952) of egg production in the bed bug. Khalifa observed that in one case spermatozoa were found in what he designates as the accessory lobe of the testes. It is surprising that spermatozoa should be found there even in one case.

HISTOLOGY

The walls of the insect testis usually consist of a peritoneal sheath enclosing the testicular tubules. The wall of the testicular tubule consists of an epithelial sheath. In the bed bug these two tissues are combined into a simple sheath of peritoneal epithelium (fig. 19, *PE*) with partitions of this epithelium dividing the testis into the seven tubules. A basement membrane is formed on the outer surface of the peritoneal epithelium enclosing the testis and on the inner surface of the testicular tubules.

The various zones of development described in the testicular tubules of many insects are not clearly distinguishable in the bed bug. The region of the tubule of the bed bug may be more conveniently designated as consisting of an apical ***germarium*** (fig. 19, *Grm*) followed by a series of ***sperm cysts*** (fig. 19, *SpC*) in various stages of development.

The germarium is a rather small mass of tissue consisting of a compact group of primary spermatogonia. In the germaria of many insects there is a large specialized cell generally believed to have a trophic function and referred to as the ***apical cell***. This cell is apparently not present in the germaria of *C. lectularius*. In a comparative study of the apical cell of insects Carson (1944) notes that in a long series of cytological studies of the spermatogenesis in Heteroptera apical cells have been found only in a very few instances. More recently Bonhag and Wick (1953) have found an apical cell complex in the lygaeid *O. fasciatus*. A thorough survey to determine in which families of Heteroptera apical cells are found might be of value as an indication of phylogenetic relationships.

Groups of primary spermatogonia become separated from the germarium and form into spherical clusters. These groups of cells become enclosed by several cells which form the wall of the sperm cyst. The nuclei of the cyst cells are characteristically large and finely granular in stained preparations.

As the sperm cells develop the cyst becomes much larger and moves down along the wall of the testicular tube (fig. 19, *SpC*). The sperm cells of any one cyst are all approximately in the same stage of development. After the spermatid stage is reached the sperm tails begin to develop, causing the sperm cyst to become elongated. At this time the nuclei of the sperm cells become aggregated into a cluster and also begin to elongate (fig. 19, *a*). They are directed toward the apex of the testicular tubule and become arranged into a compact bundle.

With the elongation of the sperm cells the cyst becomes displaced toward the center of the testicular tubule. At this time the sperm heads at the apex of the cyst are directed toward the germarium. As the cyst continues to elongate the apex of the cyst turns and begins to move toward the vas efferens (fig. 21, *SpC*). By the time the cyst reaches the vas efferens it has completely turned around and the spermatozoa are fully matured (fig. 19, *b*). The sperm cyst then ruptures and the spermatozoa pass through the vas efferens into the vas deferens and then into the seminal vesicle. This process of elongation and turning of the sperm cysts is similar to that described in the coreid *Leptocoris trivittatus* (Say) by Payne (1934) who suggests that it may be of general occurrence in Hemiptera.

In the sperm cyst containing spermatozoa which are nearly mature large spherical bodies which stain intensely with iron hematoxylin may be seen. These bodies give a negative Feulgen reaction. They are probably formed by the cast off cytoplasm of the developing sperm cells. Similar inclusions in the sperm cysts of various pentatomids have been described by Bowen (1922).

There is no outstanding difference between the histology of the vasa deferentia and the seminal vesicles. However, the muscularis of the vasa deferentia is relatively thicker. The walls of of these ducts consist of an inner layer of simple epithelium (fig. 20, *Ept*) which is surrounded by network of muscle fibers extending in various directions (fig. 20, *Mcl*). As in many insects the heads of the spermatozoa are imbedded in the epithelial lining of the seminal vesicles and their tails extend anteriorly in a spiral into the lumen. The epithelium does not appear to be secretory.

The lumina of the mesadenia are rather small and their borders are not well defined. The cells are large and their boundaries are indistinct. The cytoplasm is rather basophilic and the nuclei are moderately large and spherical.

The wall of the mesadenial reservoirs is formed by a single layer of epithelial cells. These cells can withstand considerable stretching and become very thin when the reservoir is distended. After copulation when the reservoir has been partially emptied the cells of the wall contract and are

much thicker. The boundaries between the cells are not well-defined and the nuclei are found closely associated in pairs. The epithelium of the mesadenial reservoirs is enclosed by a very sparse network of muscle fibers. This musculature probably serves to force the contents of the reservoirs into the ejaculatory pump during copulation.

THE EJACULATORY PUMP

Posteriorly the muscularis of the vasa deferentia abruptly thickens to form a bulbous muscular structure, the *ejaculatory pump* (fig. 7, *EjP*). The thick walls of this structure are formed primarily of muscle fibers which run in all directions. The ends of the vasa deferentia and ducts from the mesadenial reservoirs extend as a single blunt process into the atrium of the ejaculatory pump (fig. 7). The walls of this process are formed by muscles running concentric to the lumen of the ducts. Posteriorly the atrium of the ejaculatory pump opens broadly into the *ejaculatory duct* (fig. 7, *DE*).

Frequently when a mature male is dissected rapidly in normal saline the movements of the ejaculatory pump may be observed. These movements consist of a series of pulsatile contractions. When the muscles of the pump are relaxed the muscles of the vasa deferentia and mesadenial reservoirs contract slightly forcing spermatozoa and seminal fluid into the atrium of the pump. The constrictor muscles at the tips of the vasa deferentia and ducts of the mesadenial reservoirs probably close these ducts as the pump contracts and the semen is forced through the ejaculatory duct and phallus.

The Female Reproductive System

GENERAL MORPHOLOGY AND TERMINOLOGY

The paired ovaries of the bed bug (fig. 17, *Ov*) each consist of seven tubular chambers called *ovarioles* (fig. 17, *Ovl*). A small cord of tissue, the *apical filament* (fig. 17, *AF*) extends anteriorly from the tip of each ovariole. The apical filaments of the ovarioles merge into a single *suspensory ligament* (fig. 17, *SL*) which runs anteriorly to its attachment and, as the name implies, suspends the ovary in the abdomen.

The ovarioles are of the acrotrophic type. That is they consist of an apical trophic portion designated as the *end chamber* (fig. 17, *EC*), followed by the *germarium* (fig. 17, *Grm*) which is in turn followed by the *vitellarium* (fig. 17, *Vtl*) in which the ova mature. A cytoplasmic cord extends from the nurse cells of the end chamber through the germarium to each of the maturing ova.

In general the ovaries of the bed bug are typical of those found in heteropterans. Acrotrophic ovarioles are found only in Heteroptera and certain Coleoptera. According to the survey made by Carayon (1950) the number of ovarioles in the ovary of Heteroptera varies with different families but seven are quite frequently found and this number seems to be typical of Cimicidae and related families.

Posteriorly each ovariole narrows into a short tubular portion, the *pedicel* (fig. 17, *Pd*). The pedicels of each ovary converge and join the *lateral oviduct* (fig. 17, *OdL*). The lateral oviduct from each ovary passes posteriorly and joins the very short *common oviduct* (fig. 17, *OdC*).

At the base of each lateral oviduct near its union with the common oviduct there is an ear-like outgrowth of tissue (fig. 17, *SR*). These organs are designated here as the *sperm reservoirs*. They will be more fully discussed below.

The common oviduct opens broadly into the genital chamber (figs. 14, 17, *GCh*). There is not a clear boundary between them. The wall of the genital chamber is formed by a broad, flat membrane extending between the gonocoxopodites of the eighth and ninth segments.

In insects there is typically a spermatheca opening into the genital chamber just posterior to the common oviduct. This structure is lacking in the bed bug. Likewise an accessory gland commonly opens into the genital chamber of insects posterior to the spermatheca. This gland also is absent in the bed bug. In several heteropterans median or paired patches of tissue on the roof of the genital chamber which appear to be glandular have been described (Larsen, 1938, Ludwig, 1926, Bonhag, 1953 and Davis, 1955). No structure similar to these is found on the genital chamber of the bed bug.

The mycetome of the female is found in the vicinity of each ovary (fig. 17, *Mct*). There is not a strong connection of this organ to the ovaries as there is to the testis, but instead the mycetomes of the female are joined to a nearby lobe of the fat body.

HISTOLOGY

The ovarioles of each ovary are loosely bound together by a network of connective tissue fibers and tracheoles. Each ovariole is enclosed in a *peritoneal sheath* (fig. 18, *PS*) which also extends anteriorly from the end chamber to form a part of the apical filament (fig. 18, *AF*). At the apex of the end chamber there is a series of flattened cells which form the base of the apical filament (fig. 18, *a*). These cells continue anteriorly to form the core of the apical filament. Beneath the peritoneal sheath there is a layer of scattered cells with uniform cytoplasm and spherical nuclei (fig. 18, *b*). These cells are best seen in sections of the ovarioles of the young female. They occur mostly beneath the peritoneal sheath around the end chamber and at constrictions between developing eggs. In the end chamber the nurse cells are arranged around an acellular core (fig. 18, *c*) which posteriorly gives rise to the *nutritive cords* which pass through the germarium to the

developing eggs (fig. 18, *NtC*). In fixed material this core and the nutritive cords have a fibrous appearance.

As in other Heteroptera the nurse cells may be conveniently divided into several groups according to the size of the nuclei. At the anterior part of the chamber there is a distinct group of small cells with small nuclei which are probably diploid. The cytoplasm of these cells does not appear to merge with the acellular core as does the cytoplasm of the other nurse cells (figs. 18, 26, *1*). The nurse cells of the median portion of the end chamber are large and multinucleate. Centrally processes from the cells gradually merge with the core of the chamber (figs. 18, 26, *2*). The nuclei are large and probably polyploid. They tend to occur in clusters. At the base of the end chamber the cell boundaries are indistinct but the cytoplasm also merges with the core. In this region giant polyploid nuclei are found (figs. 18, 26, *3*).

Certain nurse cells in the median and basal portions of the end chamber are found which appear to be in a process of drastic change (figs. 18, 27, *d*). These cells are usally located toward the periphery and their cytoplasm does not extend into the core of the end chamber. The nuceli of these cells appear to be in various stages of a process leading to the formation of a dense, dark staining sphere. The cytoplasm is very basophilic and gives a positive Feulgen reaction as do the modified nuclei during all stages of their change.

Bonhag (1953) has recently described certain nuclei in the end chambers of *O. fasciatus* which pass into the core of the end chamber and which eventually disintegrate. He has termed these the cast nuclei. No nuclei such as these have been found in the end chambers of the bed bug ovaries. A cytochemical study of the changes occurring in the nurse cells of the ovary of the coreid *Acanthocephala bicoloripes* has been made by Schrader and Leuchtenberger (1952). These authors have concluded from their investigation that as nurse cells are proliferated from the apex of the end chamber and move posteriorly, there is a fusion of the cells and their nuclei alike, thus giving rise to the large nuclei in the posterior part of the end chamber. These authors also found that desoxyribonucleic acid (DNA) is extruded in the form of droplets from the nurse cell nuclei and that after certain alterations this nuclear material along with certain cytoplasmic materials passes into the yolk of the developing eggs. This extrusion of DNA constitutes an exception to the constancy of the amount of DNA found in nuclei. In any given species the quantity of DNA in the nucleus generally corresponds with the ploidy of the nucleus.

The processes of proliferation and fusion of nurse cells and nurse cell nuclei described in *A. bicoloripes* by Schrader and Leuchtenberger have also been noted by Bonhag and Wick (1953) in their study of *O. fasciatus*.

The end chamber of the bed bug differs in certain respects from that of the coreid studied by Schrader and Leuchtenberger so it is difficult to assume that all of the conclusions of these authors regarding the material that they studied apply equally well to the bed bug. The most important difference is found in the undifferentiated nurse cells at the apex of the end chamber which in the bed bug (fig. 26, *1*) are never seen undergoing mitosis as do these cells in *A. bicoloripes*. Histological sections have been made of the end chamber of the bed bug at all stages in the reproductive processes of the adult female and no mitotic figures have ever been found.

It therefore appears that the process of formation of active nurse cells as has been described in *A. bicoloripes* and *O. fasciatus* may not be entirely the same in the bed bug. There are several possibilities which may explain how this process occurs in the end chamber of the bed bug. It may be that the mitotic rate of the undifferentiated nurse cells is so low that mitotic figures are seldom encountered. However, since eggs are produced at a high rate in the bed bug one would expect the end chambers to be relatively as active as those of other Heteroptera.

A second possibility is that the female when she becomes an adult is provided with enough undifferentiated nurse cells to supply the end chamber during the adult life span so that cell proliferation is unnecessary. The possibility of this being the case was checked by comparing the amount of undifferentiated nurse cell tissue in a young virgin female with that of a female which had been producing eggs for two months. No conclusive difference in the amount of this tissue was found. Another possibility is that endomitosis of the nurse cell nuclei plays a dominant role in the increase in nuclear size. No evidence was found which either supported or negated this possibility.

The end chamber of the bed bug also appears to differ somewhat from that of *A. bicoloripes* with regard to the extrusion of DNA from the nurse cell nuclei of the latter. In the bed bug rather than the extrusion of DNA certain nuclei apparently condense into a dense sphere which gives a positive Feulgen test for DNA (fig. 27, *d*). The entire cytoplasm of the cell in which this nuclear change has occurred also gives a positive Feulgen test for DNA. On the other hand Schrader and Leuchtenberger note that in the cytoplasm of the nurse cells of *A. bicoloripes* the extruded DNA is rapidly modified so that it no longer gives a positive Feulgen test.

The interpretation of the significance of a positive DNA test of the cytoplasm of certain nurse cells of the bed bug is complicated by the fact that the symbiotes of the bed bug occur in the end chamber where they are transmitted to

the yolk of the developing eggs (Buchner, 1923). These symbiotes have been found in this study to give a positive Feulgen reaction in the cytoplasm of the mycetomes. Therefore it is possible that those nurse cells in which the cytoplasm gives a positive Feulgen reaction do so because of an abundance of symbiotes. For this reason the bed bug is not particularly suitable for the cytochemical study of the possibility that certain yolk materials are formed from DNA extruded from the nurse cell nuclei.

Immediately posterior to the end chamber is the *germarium* (figs. 18, 27, *Grm*). In the germarium the nuclei are very compact so it is difficult to make out the cell boundaries except at the periphery. In the anterior part of the germarium the germ cell nuclei may be seen in the various stages of meiosis. At least one or two oocytes (fig. 18, *Oc*) in the germarium are joined to the end chamber by a nutritive cord (fig. 18, *NtC*) and yolk is being deposited in them. The nuclei of these oocytes are in an arrested early prophase and become very large and diffuse.

The posterior part of the germarium is composed of a compact, undifferentiated tissue which is destined to form the follicular epithelium of the developing egg in the vitellarium. This undifferentiated tissue is termed the *prefollicular tissue* (fig. 18, *PfT*) and as the young oocyte develops and passes down the ovariole, a layer of this tissue forms around it. During the early stages of the formation of the yolk, nuclei of the follicular epithelium become larger, probably by undergoing endomitosis. At this time they also undergo fragmentation into two equal halves. When the egg is still relatively small the follicular epithelium may be several layers of cells thick and the nuclei in the follicular cells are oriented one on top of the other. As the egg increases in size the follicular epithelium becomes stretched out into a single layer of cells and as the cells are flattened the paired nuclei come to lie side by side (fig. 25).

In whole mounts of the follicular epithelium made after it has become a single layer of cells, one can often see that the paired nuclei fall into two distinct classes according to their size (fig. 25, *a*, *b*). The paired nuclei of the larger group apparently have undergone endomitosis at least one more time than the paired nuclei of the smaller group. Occasionally one finds a follicular cell nucleus which has not divided (fig. 25, *c*).

Nuclear fragmentation, or amitosis, apparently is not uncommon in insect tissue and there are many descriptions of it in the literature. The so-called amitotic division of the nuclei of follicular cells of the ovaries has been described in many insects and is probably of almost general occurence in this tissue.

As the egg develops and moves down the ovariole a group of follicular cells precede it and form a plug at the anterior end of the pedicel (fig. 18, *EP*). This group of cells is termed the *epithelial plug*. When the egg is fully mature this plug degenerates, allowing the egg to pass through the pedicel into the oviduct. The formation of the epithelial plug in the bed bug is much the same as it is in insects generally. However, in the bed bug it appears to have a more specialized structure than is generally found in other forms. It consists of two parts separated by a median constriction. The part of the plug adjacent to the developing egg consists of several layers of flattened cells. This is similar to the epithelial plug generally found in insects. The posterior portion of the plug is a spherical mass which projects into the lumen of the pedicel. As this part of the plug develops the cell boundaries disappear and the nuclei aggregate in the center of the cytoplasmic mass. The nuclei are of a moderate size and are at first spherical or ovate but become irregular and crumpled in appearance. In the mated female the spermatozoa accumulate in considerable numbers in the peripheral mass of cytoplasm of this part of the epithelial plug. This unusual structure may be in some way an adaptation associated with the migration of the spermatozoa to the ovarioles.

The pedicel consists of a simple tube of elastic epithelium which joins the egg plug anteriorly and opens into the lateral oviducts posteriorly. At the base of the pedicel the wall is thickened to form an unusual collar-like structure. The significance of this structure was not determined.

The wall of each lateral oviduct consists of a simple layer of elastic epithelium (fig. 15, *Epl*) surrounded by a well-developed sheath of anastomosing muscle fibers (fig. 15, *Mcl*). By making whole mounts of the flattened oviduct it is possible to see certain important histological details hitherto undescribed. In such preparations it can be seen that the epithelial cells are binucleate (figs. 23, 28) and the cell boundaries, though indistinct, may be made out. The tissue somewhat resembles the follicular epithelium but the nuclei and nucleoli are much smaller and the cytoplasm is much less basophilic. It was not determined how these cells come to be binucleate but it is quite possible that it is by fragmentation of single nuclei.

In whole mount preparations a second type of cell in the epithelium may be distinguished. These cells are long and branched and form a network extending through the epithelium (figs. 15, 23, *SpT*). The nuclei of these cells are ovate and about the same size as the nuclei of the binucleate epithelial cells (fig. 28, *a*). In individuals which have mated it may be seen that these cells contain streams of spermatozoa migrating toward the ovarioles. Often spermatozoa may be seen lying around the nuclei of these cells. It is apparent that this network of cells serves as an intracellular pathway for the sperma-

tozoa. These cells, described herein for the first time, are designated as the *sperm tract cells*.

In his study of the migration of the spermatozoa up the oviducts Cragg (1920) noted that the spermatozoa appear to move in bundles and pass inter- and intracellularly up the oviduct wall. These bundles observed by Cragg probably are in reality sections through the sperm tract cells at a time when they contained spermatozoa.

THE SPERM RESERVOIRS

The organs designated in this paper as the sperm reservoirs are the spermathecae of Berlese (1898), Cragg (1920), and others, the *Resorptionsorganen* of Abraham (1934), and the *reservoires spermatique* of Carayon (1952a). The name adopted here is based on that of Carayon.

The wall of each sperm reservoir consists of a layer of simple epithelium (fig. 29, *Ept*). Internally the reservoirs contain a mass of cells similar in all respects to the cells of the organ of Berlese and which are thus also designated as the hemocytoid cells (fig. 29, *HcC*). There are intercellular spaces among the cells but the organs have no permanent lumen. The reservoirs are joined to the lateral oviducts by a narrow solid tissue bridge.

The sperm reservoirs receive and store the spermatozoa migrating from the organ of Berlese. They are clearly not homologous to the actual spermatheca of other insects. This fact was recognized by Cragg and subsequent investigators. The true spermatheca arises as a median invagination from the ectodermal wall of the genital chamber just posterior to the common oviduct. In heteropterans in which it is present it is typically a long tubular organ consisting of a terminal glandular portion whose secretion passes into a median chamber which opens into the genital chamber by way of an efferent duct. The entire lumen is lined with cuticle. The sperm reservoirs of the bed bug are mesodermal in origin, paired, and have no lumen. There is no vestigial structure in the bed bug which could be considered to be homologous with a spermatheca.

It is interesting to note the modification of the function of the spermatheca in families of Heteroptera somewhat related to Cimicidae. Judging from the work of Carayon (1952a) the true spermatheca is apparently lacking in Anthocoridae as it is in Cimicidae. Furthermore, as in Cimicidae, special means of insemination into the hemocoel and special mesodermal organs to receive and store the spermatozoa have evolved.

In Nabidae and Miridae (Kullenberg, 1947 and Davis, 1955) true spermathecae are found but they do not function in sperm storage. Instead a special ectodermal bladder-like organ, the seminal depository, has evolved for the storage of spermatozoa. The normal mode of copulation is found in these two families although Carayon (1952b) has found that spermatozoa are found in the body cavity of nabids of the genus *Prostemma*. The reduviids are apparently similar to mirids and nabids. Gallaird (1935) has described paired ectodermal organs arising from the genital chamber of *Triatoma* and *Rhodnius* and has designated these as spermathecae. These however are not true spermathecae but are probably more related to the seminal depository of mirids. However a true spermatheca is found in these reduviids although it does not serve to store spermatozoa. Gallaird mistakenly designated this organ as the female accessory gland although he may be correct in suggesting that it produces the egg cement.

Unfortunately little is known of the reproductive system of members of the family Polyctenidae. This is a small family of rare bat parasites closely related to Cimicidae. The only study of the reproduction of this insect was made by Hagan (1931) who described its viviparous development. His study is not complete enough to determine whether or not a true spermatheca is present. However, he did not find organs comparable to the organ of Berlese or to the sperm reservoirs of cimicids. Since he did find masses of spermatozoa in the hemocoel it seems probably that insemination and the storage of the spermatozoa are at least similar to that of the bed bug.

FATE OF THE SPERMATOZOA IN THE FEMALE

Insemination and the migration of spermatozoa through the organ of Berlese have been described above. In brief the semen is injected into the organ of Berlese where the spermatozoa pass to the periphery of the organ. From here they pass into the body cavity.

By dissecting and sectioning females a known time after mating, the course of the spermatozoa in the female may be traced. In two to four hours after mating the spermatozoa appear in large numbers in the hemocoel and may be seen to congregate around the sperm reservoirs. After about four hours most of the spermatozoa have passed into the sperm reservoirs and lie at random in masses among the hemocytoid cells (fig. 24, *SpM*). After several days in the sperm reservoirs, the spermatozoa become oriented into several arc-shaped bundles. The spermatozoa of these bundles lie parallel to each other and head in the same direction (fig. 29, *SpM*). The spermatozoa are stored in this condition and gradually move out through the tissue bridge and up the walls of the lateral oviducts by way of the sperm tract cells. It could not be determined whether or not the sperm tracts extended into the pedicels but the spermatozoa can definitely be seen in the wall rather than the lumen of the pedicel. After passing up the pedicel the spermatozoa accumulate in the lower portion of

the epithelial plug. Some do pass through the epithelial plug and are seen widely scattered between the follicular epithelium and the connective tissue sheath. The fertilization of the egg will be discussed below.

The migration of the spermatozoa out of the sperm reservoirs is largely influenced by the taking of a blood meal by the bug. If a whole mount is made of the oviduct of a mated female about twenty hours after feeding, one finds that the cells of the sperm tracts are heavily congested with spermatozoa passing toward the ovary (fig. 23, *SpT*). Whole mounts made on successive days after feeding show progressively fewer spermatozoa in the sperm tracts. About a week after feeding only a few isolated spermatozoa are found in the oviducts (fig. 28, *SpT*). If the insects are then fed again the spermatozoa reappear in the sperm tracts of the oviducts. It is thus seen that a wave of spermatozoa passes up the oviducts to the ovaries following each meal.

The production of eggs by the bed bug is similarly correlated with feeding. In this investigation the bugs were fed once a week. About two days after feeding the first eggs are laid and laying continues for about five days after feeding and then stops until the bug obtains another meal. The peak of egg-laying occurs on the third day after feeding. As has been pointed out before, eggs are laid only by females that have mated.

The consequence of the correlation of the sperm migration with feeding and egg production is that spermatozoa reach the ovaries in abundant numbers when eggs are being produced and are ready for fertilization. On the other hand the spermatozoa do not migrate to the ovaries when egg production stops and so they are not needlessly expended.

In the case of females isolated after a single mating and fed once a week egg-laying continues usually for about seven weeks. During the last week most of the eggs laid are sterile. Mellanby (1939) has shown that this period of fertility from a single mating is the same whether the female receives a large amount of sperm or only a moderate amount, and that the period is the same whether the female is fed frequently and produces a large number of eggs or fed infrequently and produces relatively fewer eggs. Therefore he concludes that the duration of the period of fertility from a single mating is related to the duration of viability of the spermatozoa rather than to the exhaustion of the spermatozoa. In the present investigation mated females isolated for seven weeks were sectioned and spermatozoa were found intact in the sperm reservoir. Since egg production in these females had stopped these spermatozoa were probably not viable. This evidence gives weight to Mellanby's belief that length of the period of fertility is related to the length of the viability of the spermatozoa.

In his study of the fate of the spermatozoa in the female bed bug Abraham (1934) concluded that the function of the organs referred to here as the sperm reservoirs is primarily to resorb the excess spermatozoa. He reports that the spermatozoa are broken down in these organs to a granular detritus in two to three weeks and eventually are resorbed. He therefore designated these organs as the *Resorptionsorganen*. Abraham's conclusions are in apparent support of Berlese's theory of hypergamesis (Berlese, 1898).

In the present investigation no evidence was found indicating that the sperm reservoirs resorb spermatozoa to the extent described by Abraham. On the contrary, as has been indicated above, it is the conclusion of this study that these organs function primarily in the maintenance of the spermatozoa. Bundles of intact spermatozoa have been observed in histological sections of the sperm reservoirs long after the period in which Abraham stated that the spermatozoa were disintegrated. Another fact that contradicts Abraham's conclusion is that a single mating lasts the female about seven weeks. It is therefore reasonable to expect that the spermatozoa are stored somewhere in the body of the female during this time. The only place they are found in numbers for a long period of time is in the sperm reservoirs.

It is quite likely that defective or dead spermatozoa are phagocytized in the sperm reservoir as well as elsewhere in the body of the female. However there was no evidence found in the present study that normal active spermatozoa are destroyed in large numbers except as a normal result of most of them having failed to successfully fertilize an egg after migrating to the ovaries. This is the fate of all but a very small fraction of the spermatozoa of the bed bug as well as of the spermatozoa of animals in general. The impressively large quantities of sperm which the female receives probably reflect the rather difficult route that the spermatozoa must travel. If this fact is recognized, there seems to be no need to seek an accessory function such as hypergamesis to account for the excess sperm.

In instances in which phagocytosis can be observed this activity does not necessarily indicate that the spermatozoa or seminal fluid is being used for nutritive purposes (i.e., hypergamesis) any more than this would be indicated by the phagocytosis of any foreign material. In other words the final result of the phagocytic activity may very well be the breakdown of the material into substances which may be excreted rather than utilized by the organism.

EGG PRODUCTION AND FERTILIZATION

The end chamber rather than the undifferentiated follicular epithelium appears to form the yolk during the early stages of the formation of the egg. At this time the yolk is very finely

granular and only slightly basophilic. The very large egg nucleus is centrally located in the yolk mass (fig. 18, *Oc*).

The secretion of yolk material by the follicular epithelium appears to begin by the time that follicular cells have been formed into a single layer around the egg. By this time the follicular cells are binucleate, the nuclei and nucleoli have increased in size, and the cytoplasm has become more abundant and more basophilic. Yolk granules can be seen forming at the free margin of the follicular epithelium. These granules appear to coalesce forming the larger yolk granules which are dispersed among the yolk formed during the earlier stages. These granules are very basophilic (fig. 30).

As the egg becomes larger and more elongated, the nucleus comes to lie in the posterior one-third of the egg and to one side next to the follicular epithelium (fig. 18, *En*). The connection to the end chamber by way of the nutritive cord is lost, and the remainder of the yolk is formed entirely by the follicular epithelium.

Only one egg at a time is formed in each ovariole and the ovarioles of each ovary form the eggs in rotation so that at any one time there is not more than one fully formed egg in each ovary.

Mellanby (1939) and previous workers have mentioned that there is no development of eggs in virgin females. This idea is not entirely correct. Eggs are formed up to a certain stage whether or not the female has mated. In the virgin female the egg yolk is formed until the egg is about two-thirds of its full size. If after about a week the egg has not been fertilized, resorption of the yolk begins. In the early stages of resorption the follicular epithelial cells invade the yolk and the amount of yolk gradually diminishes. In the final stages of resorption only the follicular cells remain, and these form a small mass which eventually breaks down into granular detritus. In the female which has been kept isolated as a virgin for several weeks each ovariole may be seen to contain a series of eggs in progressive stages of resorption. However, the rate of egg production in the virgin female is probably not as high as in the mated female. It should be noted that the resorption of unfertilized eggs by the virgin female probably conserves food material and may therefore be of survival value.

In the mated female the follicular epithelium begins to form the chorion at about the same time that the blastoderm of the embryo is formed (fig. 30, *Bld*, *Ch*). At this time the cytoplasm of the follicular epithelium becomes very much more basophilic and the inner surface of the tissue becomes smoother (fig. 30, *FE*). The chorion as it is formed is at first intensely stained by hematoxylin but later this basophilia is lost and the chorion is seen as a clear refractile layer. The development of the embryo continues as the chorion is formed and by the time that the egg is laid the embryo is usually in the early stages of blastokinesis.

Although the fusion of the male and female pronucleus has not been seen in this or previous investigations it is certain that fertilization occurs in the ovary and before the chorion is formed. A study of serial sections of mated females shows that the egg nucleus remains unchanged until the egg is well developed. Therefore the fusion nucleus is not formed until the egg is almost completely developed. The penetration of the spermatozoa probably also does not occur until the egg is well developed since spermatozoa are not found in the follicular epithelium until this time.

After the egg has passed into the oviduct the follicular epithelium and epithelial plug break down and begin to disintegrate. In various insects this small mass of disintegrating tissue is yellow and has therefore been called the *corpus luteum*. This structure, however, is in no way comparable to the *corpus luteum* of mammals.

The manner in which mating initiates the complete formation of eggs has been studied by Mellanby (1939). By eliminating other factors Mellanby has concluded that the presence of spermatozoa in the female is responsible for initiation of oviposition. How the spermatozoa effect this stimulation has not been determined but this problem is now being studied by the author. As an aid to this study a successful technique of artificial insemination has been developed. Descriptions of this technique will appear in an anticipated publication concerning ovarian stimulation in the bed bug.

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SUMMARY

1. The morphology of the abdomen and of the male and female genitalia is described and compared with other heteropterans. Although the genitalia are highly modified, most of the basic elements of the generalized heteropteran genitalia may be distinguished. The genital muscles and their actions are also described.

2. The structure and evolution of the organ of Berlese is discussed. Its primary functions are phagocytosis, prevention of bleeding, and prevention of damage to the internal organs during copulation.

3. The anatomy and histology of the male reproductive organs and ducts are described.

The testes are similar to those of other Heteroptera. No apical cell is found in the germarium. The accessory glands are the mesadenial type.

4. The anatomy and histology of the female reproductive organs and ducts are described. True spermathecae and accessory glands are lacking. The spermatozoa are received and stored in the sperm reservoirs. The formation of giant nuclei and the liberation of DNA by the ovarian end chambers appears to be different from what has been described in certain other Heteroptera. The cells of the follicular and lateral oviduct epithelium are binucleate. The lateral oviducts contain a network of special cells through which the spermatozoa migrate to the ovarioles.

5. Eggs are partly formed then resorbed if insemination has not occurred. Spermatozoa migrate to the ovarioles only after the insect has fed. The egg is fertilized after most of the yolk has been deposited. The chorion and blastoderm are formed at about the same time.

6. Artificial insemination has been successfully performed by injecting semen into the organ of Berlese.

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PROLONGED SURVIVAL OF INSECT TISSUES IN VITRO[1]

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The epidermis of insects has long served the student of growth as excellent material for the solution of morphogenetic problems. Unfortunately, as others have recently pointed out (Schmidt and Williams, 1953; Goodchild, 1954; Grace, 1954) no one has succeeded in culturing insect epidermis (or any other insect tissues for that matter) to the degree that vertebrate tissues have been cultured. Insect blood has often been used as a culture medium but it is surprising that it (with or without cells) is an unsatisfactory medium for all insect tissues save the spermatocytes and ovaries (Schmidt and Williams, 1953; W. H. Telfer, unpublished observations). Thus Goodchild (1954) has described changes which appear to be degenerative in fragments of *Rhodnius* epidermis maintained in hanging drops of *Rhodnius* blood for 10 to 14 days. Similarly in our laboratory we have found that although immature spermatocytes of the cynthia moth, *Samia cynthia* (Drury), and the polyphemus moth, *Antheraea polyphemus* (Cram.), undergo spermatogenesis in the blood of developing adult silkworms (to which phenylthiourea[2] and streptomycin were added), fragments of pupal wing epidermis underwent prompt degenerative changes similar to those described by Goodchild: they tended to form rounded granular masses and the nuclei became indistinct within a few days. After five to six days all noticeable changes ceased. Cultures maintained in Ringer's solution (Ephrussi and Beadle, 1936) lasted somewhat longer but degenerated after about 10 days. However, by using a chemically defined medium (TC 199) originating in the laboratories of Morgan, Morton, and Parker (1950)[3] we have maintained epidermis from the pupal wing and antenna alive *in vitro* for between 20 and 35 days.

Diapausing pupae were surface sterilized by immersion in a solution of 0.05% $HgCl_2$ in 50% ethyl alcohol for 1 minute, and then rinsed in two changes of sterile distilled water. They were dissected in sterile insect Ringer's solution. Pieces of wing and antenna were excised, placed in a small drop of Ringer's solution to which a few crystals of phenylthiourea and streptomycin had been added and cut into fragments approximately 0.5 mm. square. These fragments were then placed in drops of TC 199 (to which phenylthiourea and streptomycin had been added as previously), on glass cover slips and sealed onto depression slides with melted paraffin. These hanging drop cultures were maintained at 25° C. and the medium was renewed approximately every five days. Altogether 148 cultures were prepared in this manner and examined frequently under a phase contrast microscope. Periodically representative cultures were fixed and stained with hematoxylin and eosin.

In TC 199 the wing epidermis and, to a lesser extent, the antennal epidermis attaches to the cover slip and spreads peripherally. The thicker center of the fragment thus becomes surrounded by a progressively thinning cytoplasmic network throughout which nuclei are scattered (fig. 1).

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[2]Following the suggestion of Schmidt and Williams (1953) phenylthiourea (twice recrystallized from hot ethanol) was added to inhibit the tyrosinase present in insect blood and thus prevent the formation of toxic quinones.

[3]Medium available as T. C. Medium 199 from Difco Laboratories, Detroit 1, Michigan. The use of this medium was suggested by Dr. William E. Beckel of the Defense Research Board of Canada.

This spreading does not occur to the same extent over the periphery of the fragment to produce the rounded colony of cells often observed in cultures of migrating fibroblasts, but it happens consistently. Most of the spreading occured within 24 hours after preparation but subsequent day to day changes were observed and recorded by drawings and ocular micrometer measurements. Disturbances of the culture such as those occurring during subculture commonly induced transitory but vigorous spreading activity. However as the culture aged this spreading

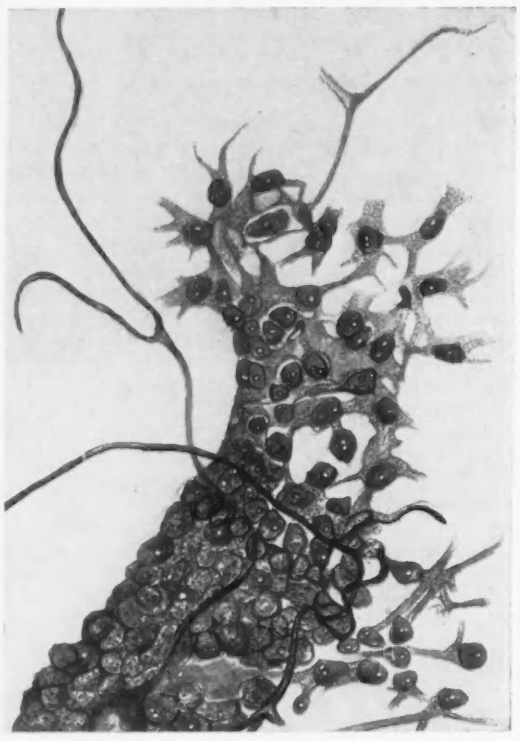

FIG. 1. Spreading portion of a 10-day old culture of epidermis from the pupal wing of *Antheraea polyphemous* maintained in medium TC 199. Minute tracheoles can be seen running through the fragment and out at the left; the elongated process at the top right is a cytoplasmic extension. Note the prominent nucleoli present in all nuclei. (Phase contrast, camera lucida X430).

response progressively lessened so that a 15 day culture spread about half as much as a 10-day culture and a 20-day culture spread only slightly.

Since the nuclei are spread thinly they are easily observed and provide an index to the condition of the culture; in a fresh, healthy culture most of the nuclei are clear save for one or two prominent nucleoli, while in a degenerate or aged culture the nuclei are extremely granular and the nuclear membranes become indistinct. As was expected no mitotic figures were observed in any of the cultures, for in the absence of the growth hormone of the prothoracic glands pupal epidermal cells do not divide even after extensive wounding (Smith and Schneiderman, 1954). Cultures of wing and antennal epidermis from animals midway in adult development ceased differentiating when placed in TC 199 but survived as long as the diapausing tissues (cf. Ketchel and Williams, 1953). As soon as the recently crystallized prothoracic gland hormone (Butenandt and Karlson, 1954) is made available it should be possible to maintain growing cultures of epidermal cells using the technique just described, and perhaps supplementing the maintenance medium. Although TC 199 is certainly not the "final word" in insect tissue culture or even insect tissue maintenance, its use may facilitate experimentation in both developmental morphology and in studies of insect viruses and Rickettsial organisms.

We gratefully acknowledge the helpful suggestions of Dr. C. M. Williams, Dr. G. R. Wyatt and Dr. Henry S. Fuller who read the typescript of the present paper.

Recently by adding 2% Difco Bactopeptone we have achieved much longer survival in our cultures: epidermis, heart fragments, pericardial cells, and Malpighian tubules survived for more than 6 weeks.

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FLUORESCENCE IN COCHROACHES[1]

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We have observed hitherto unreported fluorescence in various tissues, organs, and products of 19 species of cockroaches during irradiation with ultraviolet light.

Fluorescence is a characteristic of many compounds and, by itself, may not be particularly important. However, changes occurring in the fluorescent colors of certain organs, such as accessory reproductive glands, may indicate changes in secretory activity and thus be valuable visual indicators in physiological studies.

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So far as we know, fluorescence in cockroaches has been described for only (1) "pigment-free tissue" (Metcalf and Patton, 1942); (2) the malpighian tubes (Busnel and Drilhon, 1942; Metcalf and Patton, 1942); (3) the secretion of the latero-cervical organ (Pavan, 1954b, 1954c; Pavan and Willis, 1955); and (4) the euplantulae, articular membranes, and unspecified parts of the abdominal organs (Pavan, 1954b).

Except for riboflavin in the malpighian tubes of four species of cockroaches, nothing is known of the chemical composition of the fluorescent materials found in cockroaches. Quite possibly,

TABLE I

COLOR OF THE FLUORESCENCE IN CUTICLE AND PRODUCTS OF COCKROACHES IRRADIATED WITH ULTRA-VIOLET LIGHT
Key: B = blue; G = dull yellowish green; Y = greenish yellow; DG = dark green; DY = dull greenish yellow; OG = olive green; —— = structure not present; . . . = not examined.

Family Subfamily Species	Cuticle		Products								
				Eggs		Embryos					
	Sclerotized	Unsclerotized	Oothecae	Chorions	Contents	Tissues	Gut Contents	Embryonic Membranes	Feces	Secretion of Cervical Organ	Spermatophores
BLATTIDAE											
BLABERINAE											
Blaberus craniifer Burmeister	Y	B[a]	Y	Y	Y	. . .	. . .	. . .	DY	——[a]	. . .
Blaberus giganteus (Linnaeus)	Y	B	. . .	. . .	. . .	. . .	. . .	. . .	DY	. . .	. . .
BLATTINAE											
Blatta orientalis Linnaeus	Y	B[a]	G	DY	Y	B	Y	B	DY	B[a]	B
Eurycotis floridana (Walker)	Y	B	G	DY	Y	B	Y	B	DY	B[b]	B
Neostylopyga rhombifolia (Stoll)	Y	B	G	DY	Y	B	Y	B	DY	B	B
Periplaneta americana (Linnaeus)	Y	B[a]	G	DY	Y	B	Y	B	DY	B[a]	B
Periplaneta australasiae (Fabricius)	Y	B	G	DY	Y	B	Y	B	DY	B[b]	B
Periplaneta brunnea Burmeister	Y	B	G	DY	Y	B	Y	B	DY	B	B
Periplaneta fuliginosa (Serville)	Y	B	G	DY	Y	. . .	. . .	. . .	DY	. . .	. . .
ECTOBIINAE											
Ectobius livens (Turton)	Y	B	G	DY	DG	. . .	. . .	. . .	DY	. . .	. . .
EPILAMPRINAE											
Leucophaea maderae (Fabricius)	Y	B[a]	Y	Y	B, Y	B	Y	B	DY	——[a]	B
Nauphoeta cinerea (Olivier)	Y	B	Y	Y	B, Y	B	Y	B	DY	. . .	B
Pycnoscelus surinamensis (Linnaeus)	Y	B	Y	Y	B, Y	B	Y	B	DY	. . .	. . .
PSEUDOMOPINAE											
Blattella germanica (Linnaeus)	Y	B[a]	DY	DY	OG	B	Y	B	DY	——[a]	B[c]
Blattella vaga Hebard	Y	B	DY	Y	B	B	Y	B	DY	. . .	B[c]
Loboptera decipiens Germar	Y	B[a]	. . .	. . .	. . .	. . .	. . .	. . .	DY	——[a]	. . .
Parcoblatta pensylvanica (Degeer)	Y	B	DY	Y	Y	. . .	. . .	. . .	DY	. . .	. . .
Supella supellectilium (Serville)	Y	B	DY	Y	Y	B	Y	B	DY	——[b]	B
DIPLOPTERIDAE											
DIPLOPTERINAE											
Diploptera dytiscoides (Serville)	Y	B	Y	Y	Y	B	Y	. . .	DY	. . .	B

[a]Pavan, 1954b.
[b]Pavan and Willis, 1955.
[c]Dry spermatophores dropped by females.

the greenish-yellow fluorescence in cockroach eggs is due to riboflavin; fluorescent substances, identified as riboflavin, have been found in the eggs of other insects from several orders (Busnel and Drilhon, 1942; Bodine and Fitzgerald, 1947; DeLerma, 1952).

MATERIALS AND METHODS

We examined the species of Blattaria that are listed alphabetically by subfamily (after Rehn, 1951) in table I.

All species were from cultures maintained on Purina dog chow checkers. Specimens were killed with chloroform just prior to examination or, if scarce, were collected as they died and examined while still moist.

The insects were examined at 9X magnification with long wave (3660 Å) ultraviolet light.

OBSERVATIONS

The fluorescent colors found in the cuticle and products of cockroaches are listed in Table I. The fluorescent colors found in certain internal organs are listed in Table II.

The pale greenish-yellow fluorescence of sclerotized cuticle could be seen best in newly molted insects. Except for intensity of color, the yellow fluorescence of cockroach cuticle seemed to be similar to the brilliant yellow fluorescence described by Pavan (1954, 1954a) for the cuticle of scorpions.

Oöthecae of the so-called viviparous cockroaches (Blaberinae, Diplopterinae, Epilamprinae) fluoresced very bright greenish yellow. However, the darker oöthecae of the oviparous species fluoresced much less distinctly; the oöthecae of the Blattinae fluoresced hardly at all.

TABLE II

COLOR OF THE FLUORESCENCE IN VARIOUS ORGANS OF COCHROACHES IRRADIATED WITH ULTRA-VIOLET LIGHT

Key: B=blue; P=pink; X=structure present but nonfluorescent; Y=greenish yellow; DG=dark green; OG=olive green; LY=very light greenish yellow; YW=yellowish white; —=structure not present; ...=not examined.

Family Subfamily Species	Alimentary Canal	Fat Body	Malpighian Tubes	Muscle (Coxal)	Nerve Cord	Salivary Glands		Tracheae		Ventral Abdominal Gland	Reproductive Organs					
											Male			Female		
						Glands	Reservoirs	Outer Surface	Lining[a]		Testes	Accessory Glands: Utricular	Accessory Glands: Phallic	Ovaries	Corpora lutea	Colleterial Glands
BLATTIDAE																
BLABERINAE																
Blaberus craniifer	B	B	Y	Y	LY	LY	B	Y	B	...	...	...	...	...	...	...
Blaberus giganteus	B	X	Y	LY	LY	LY	B	Y	B	...	...	...	...	...	...	...
BLATTINAE																
Blatta orientalis	B,Y	X	(Y,B)[b,c]	Y	Y	Y	B	Y	B	Y	...	B,Y	B,Y,P	Y	Y	B
Eurycotis floridana	B	X	Y	Y	Y	Y	B	Y	B	Y	...	B,Y,P	B,Y	Y	...	B
Neostylopyga rhombifolia	B,Y	X	Y	Y	Y	Y	B,YW	Y	B	Y	Y	B,Y,P	B,Y	Y	...	B
Periplaneta americana	B,Y	X	(Y,B)[b,c]	Y	Y	Y	B	Y	B	Y	Y	B,Y,P	B,Y,P	Y	Y	B
Periplaneta australasiae	B	X	Y	Y	Y	Y	B	Y	B	Y	Y	B,Y	B,Y,P	Y	...	B
Periplaneta brunnea	B,Y	X	Y	Y	Y	Y	B	Y	B	Y	Y	B	Y	Y	...	B
Periplaneta fuliginosa	B	X	Y	Y	Y	Y	B	Y	B	...	...	...	...	...	...	...
ECTOBIINAE																
Ectobius livens	Y	X	Y	Y	Y	Y	B	Y	B	...	...	...	...	DG,Y	Y	B,Y
EPILAMPRINAE																
Leucophaea maderae	B	X	Y	Y	Y	Y	B	Y	B	—	B,Y	B,Y	B,Y	LY	...	B
Nauphoeta cinerea	B,Y	X	Y	Y	Y	Y	B	Y	B	—	Y	B,Y	B	LY	Y	B,Y
Pycnoscelus surinamensis	B	X	Y	Y	Y	Y	B	Y	B	—	...	...	...	LY	...	B
PSEUDOMOPINAE																
Blattella germanica	B,Y	X	Y[b]	Y	Y	Y	B	Y	B	—	Y	B,Y	Y	OG	Y	B
Blattella vaga	B	X	LY	Y	Y	Y	B	Y	B	—	Y	B	Y	B	...	B
Loboptera decipiens	B,Y	X	Y	LY	Y	Y	B	Y	B	...	...	...	...	...	...	...
Parcoblatta pensylvanica	B	X	Y[b]	Y	...	Y	B	Y	...	...	...	...	...	Y	Y	B,Y
Supella supellectilium	B,Y	X	Y	Y	Y	Y	B	Y	B	—	Y	B	...	Y	Y	B,Y
DIPLOPTERIDAE																
DIPLOPTERINAE																
Diploptera dytiscoides	B	X	Y	Y	Y	Y	B	Y	B	—	Y	B,Y	LY	LY	Y	B

[a]Tracheal linings in exuvia.
[b]Metcalf and Patton, 1942.
[c]Busnel and Drilhon, 1942.

The uncolored anterior ends of partially formed oöthecae of *Blatta orientalis* and *Periplaneta americana* fluoresced blue, but this color disappeared as the oöthecae darkened.

SUMMARY

Fluorescence of several colors is reported for various tissues, organs, and products of 19 species of cockroaches which were irradiated with ultraviolet light. The results of these observations are summarized in Tables I and II. It is suggested that differences in the fluorescent colors of certain organs may indicate changes in secretory activity.

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OBSERVATIONS ON THE FLIGHT OF DRONE HONEY BEES[1]

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The flight activities of drone honey bees (*Apis mellifera* L.) have received little attention, although tens of thousands of queens are sold each year and each queen must be mated to one or more flying drones. This paper reports tests on the flight habits of marked drones and also on the amount of food eaten by drones, which were conducted at Baton Rouge, La., from March to October in 1952, 1953, and 1954.

METHOD

All the drones used for test flights were marked with a mixture of pigment and shellac. Tests were usually conducted under weather conditions that experience had indicated to be favorable for flight. Drones used in the afternoon, usually between 1 and 4 p.m., were caught at the hive entrance as they returned from a flight, and were marked, caged, and released within 10 minutes at known distances from the hive. The confinement of test drones even for a short period may have influenced their subsequent flight. It is certain that such drones had had at least one flight to orient themselves to the surrounding area. Drones used in forenoon flights, usually between 9 and 10 a.m., were captured within the hive; so it is possible that some had never been outside. Different lots of drones were released at distances from ½ to 2½ miles east, west, north, and south of the apiary. There were no releases beyond ½ mile south of the apiary because of the presence of the Mississippi River.

DURATION OF FLIGHT

In observations on the duration of natural flight, from the hive, the exits and returns of 20 marked drones were noted. The time spent away from the hive ranged from 1 to 57 minutes. It is presumed that the entire time was spent in flight, since drone honey bees are not known to rest during a flight. Some of the drones made four flights in one afternoon. These results are fairly close to those reported by Howell and Usinger (1933).

HOURS OF FLIGHT

The maximum number of flights took place between 2 and 3 p.m. Most drones flew only

[1]In cooperation with Louisiana State University.

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in the afternoon, although an occasional one was seen to fly between 11:30 a.m. and 12 noon. There were practically no flights after 4:30 p.m. These results are much like those reported by Minderhoud in Holland (1932). At Davis, Calif., in mid-May most of the drones flew between 2 and 4:30 p.m., according to Howell and Usinger (1933). This corresponds closely to the period 1:45 to 4.35 p.m., when virgin queens normally fly (Oertel 1940). Probably clouds, shade from nearby trees, temperature, weather on preceding days, and perhaps other factors influence the time when the peak of drone flight occurs. There is a variation from colony to colony, from day to day, and from season to season (Oertel 1940).

SPEED OF FLIGHT

The speed of flight of the first drones to return to the hive from the lots released ½ to 1½ miles away is given in Table I. The time used by these drones to get back to the hive ranged from 2 to 32 minutes. The first drone to return

TABLE I

SPEED OF FLIGHT OF RETURNING DRONES FROM RELEASE POINTS ½ TO 1½ MILES FROM THE HIVE

Year	Number of lots	Miles from hive	Minutes for first drone to return		Calculated speed, m.p.h.
			Average	Range	
1952	12	½	3.8	2–8	7.9
	18	1	10.5	6–32	5.7
	1	1½	10.0	....	6.0
1953	11	½	4.3	3–9	7.0
	6	1	9.5	6–19	7.9
1954	2	½	6.0	3–9	10.0
	4	1	9.2	5–19	6.5

in each of 11 lots released 2 miles from their hive came back in 9 to 36 minutes. Their average was 18 minutes and their rate was 6.6 m.p.h. No drones returned from 5 of these lots.

Many drones could not be expected to return. The amount of food carried, the age of the drone, natural enemies, and the possibility of death through mating all tend to reduce the numbers returning to the hive. The first drone to return may have been the fastest flier or the one most familiar with the terrain. It is assumed that he flew in the most direct line, but those that returned later may have flown off-course or in a circle until they saw a familiar landmark.

According to Schnell-Larsen (Hocking 1953), drones in a test flew at the rate of 180 cm. per second (4 m.p.h.), but it is not known whether free-flying drones were used.

[2]For additional information on the feeding of drone bees see Oertel, Emerson, and Wheeler (1953).

FOOD CONSUMED

Sixty-three drones, caged as they returned to the hive, took 0.9 gram of a 50-percent sugar sirup during the first hour, or 14.3 mg. per drone. At the end of 24 hours 7.8 grams of sirup had been taken, but 9 of the drones had died.[2] Another lot of 50 drones similarly caged took 0.57 gram of honey during the first hour, or 11.4 mg. per drone. In 24 hours 2.96 grams had been taken, but 15 drones had died. Fifty drones taken from combs within the hive and caged took 0.3 gram of honey during the first hour, or 6 mg. per drone. In 18 hours 1.6 grams had been taken, but 16 drones had died. Drones that had just returned from a flight took nearly twice as much honey as the ones taken from the combs.

The weight of drones that returned to the hive from flight averaged 185 mg., and that of young, caged drones that had not flown averaged 220 mg. Most of the additional weight probably was accumulated feces, since many of the drones defecated when they were handled. The wing area of the drone is larger than that of the worker, but in about the same ratio to body weight. Since the drone is heavier but flies at about the same speed, it is assumed that he uses more energy in flight than the worker.

The observer cannot be sure that his test drones, whether taken from the combs or at the hive entrance, have a normal amount of honey in the honey stomach. It is not known whether those that do not return run out of fuel or cannot find their way back to the hive. An effort was made to feed caged drones just prior to their release, but they became smeared with food and many did not fly when released.

Jongbloed and Wiersma (1934) calculated that a lot of worker honey bees weighing 1 kg. (about 100 mg. per bee) needs 18.6 calories of energy to fly 1 km. If we assume that the drone weighs twice as much and flies at the same speed as the worker but uses twice as much energy, then 1 kg. of drones also need 18.6 calories to fly 1 km. This amount of energy is contained in about 56 mg. of honey. Worker bees tested by Hocking (1954) by means of a flight mill could fly for 6.17 hours for a calculated distance of 57 km. on a consumption of 6.77 mg. of sugar per hour. The amount of honey taken by a caged drone indicates that the honey stomach can hold enough to permit him to fly several miles. The amount of honey that the stomach actually contains when he leaves the hive for a flight has not been learned.

PERCENTAGE OF DRONES THAT RETURNED TO THE HIVE

The percentages of drones that returned to the hive from known points of release are given in Table II. About half of those that were

released ½ mile from the hive returned, but only a few of those released 2 miles and none of those released 2½ miles away. Large differences were found between different lots of drones. For example, for the ½-mile release point there was a range from 20 to 85 percent. The direction of the release points from the apiary had no effect upon returns.

EFFECT OF VARIOUS FACTORS UPON RETURNS

Time of Day.—Since drone honey bees fly almost entirely in the afternoon, some tests were carried out to learn how many drones would return if they were released in the forenoon. Table II, shows that drones can get back to the hive nearly as well in the forenoon as in the afternoon.

Removal of Antennae.—In these tests 100 drones were used as controls, and 145 had their antennae wholly or partially removed. Thirty percent of the drones with one or both antennae removed and 40 percent of those with half of each antenna removed returned to the hive. Since only 42 percent of the unmutilated drones returned, the injury was not serious enough to prevent the return of drones to the hive.

TABLE II

PERCENTAGE OF DRONES RETURNING FROM VARIOUS DISTANCES WHEN RELEASED IN FORENOON AND IN AFTERNOON

Miles from hive	Forenoon		Afternoon	
	Number released	Percent returned	Number released	Percent returned
½	245	52.2	285	47.0
1	270	5.5	600	21.1
2	50	1.6	465	0.04
2½	....	....	125	0

Low Temperature.—The temperature at the time of the releases reported in Table II ranged from 80° to 100° F., which is within the normal flight-temperature range for drones. The effect of lower temperatures upon drone return was observed between 9 and 10 a.m. on April 13 and 14. Fifty marked drones were released ½ mile north and 50 were released 1 mile north from the hive. The sky was clear, the temperature 58° to 60° F., and the wind from the north between 5 and 10 miles per hour. Worker bees were not making flights at that time. None of the drones returned to the hive.

Do Drones Use the Sun to Help Them Find the Hive?—Worker bees use polarized light and the position of the sun in the sky as an aid in orientation when working in the field, according to von Frisch (1954). An experiment was conducted to learn whether drones use the position of the sun to help them find their hive. A hive was covered by a tent so that when the hive was opened the drones could not see the sun or the sky. Drones were caught, marked, and caged, and the cages were put into light-tight cartons under the tent. The cartons were then taken to the points where the drones were released. Since these tests were made at about 9 a.m., we can be reasonably sure that the caged drones had seen the sun and the sky only in the afternoon on previous flights. Other lots of drones were caged in the same way and then released when the sky was cloudy or partly cloudy. As shown in Table III, the drones found the hive about as well in the forenoon as in the afternoon, whether or not the sky was clear, and their inability to see the sun or sky when the hive was opened did not prevent returns. Probably drones use landmarks to find their way back to their hive.

TABLE III

NUMBER OF DRONES RELEASED AND PERCENT RETURNED UNDER CLEAR, CLOUDY, OR SEMI-CLOUDY SKIES, WHEN DRONES DID NOT SEE SUN OR SKY UNTIL TIME OF RELEASE

Miles from hive	Forenoon		Afternoon	
	Number released	Percent returned	Number released	Percent returned
		Clear sky		
½	75	32	25	32
1	50	4	25	16
		Cloudy		
½	50	42	0	....
1	50	6	0	....
2	0	....	50	8
	Clear to partly cloudy			
½	50	28	75	41
1	0	....	65	23
Total or average	275	23.3	240	25.8

HOW DO DRONES AND QUEENS FIND EACH OTHER?

We do not know how the drone and queen honey bees find each other during the mating flight—whether by sound, sight, odor, chance, or some other means, or by a combination of them. On the assumption that the drones generally fly only ½ mile from the apiary and to a height of not more than 50 feet, there are approximately 41 million cubic yards in their flying space.

Most of the flights of drones, all those of virgin queens, and the play flights of workers take place in the afternoon. The reason for this is not known, since external conditions in the forenoon often appear to be favorable for flight. Drone flights generally take place at approximately the maximum temperature and the minimum relative humidity for the day, after the light intensity has started to decline, when the sky is clear to only partly cloudy, and when wind velocity is fairly low. Probably a combination of high light intensity and suitable temperature mainly influences the time of drone flight.

It is difficult to explain, under our present knowledge of bee behavior, how the drones become aware of external conditions. The brood chamber is dark, and the temperature and relative humidity are fairly constant. Perhaps the worker bees transmit in some unknown manner the information that external conditions are suitable for flight or scout drones go out to test conditions.

"Schools of drones," loose aggregations of individuals that fly in limited areas as a means of insurance that the virgin will find a drone, were discussed long ago by Doolittle (1892). Mating activity when many individuals are flying about in a small area (swarms) is known to occur in other species of insects (Roeder 1953). However, there is little information as to the existence of groups of drone honey bees flying about in the air. Nor do we know how the queen would find a group of drones except that she might more easily see, hear, or smell a group than an individual. Since the drone honey bee has much larger compound eyes than the queen and his antennae possess many more sensillae, some investigators have concluded that the drone seeks out the queen (Imms 1931). Kruger (1951) believes that in bumble bees, *Bombus* and *Psithyrus*, vision plays a more important part than odor, although the male odor is easily recognized. It has not been established that drone honey bees have a marked odor while they are in flight.

The range of drone and virgin queen flight was studied by Donald Peer (unpublished Ph.D. thesis, University of Wisconsin) by means of the color of progeny from cordovan queens. He found at least one mating when the apiary containing cordovan virgin queens was 6 miles from the drone source. It is not known how far the queen flew to meet the drone. Possibly the nature of the terrain may affect the distance that drones fly.

Sladen (1920) reported that when he provided, on an isolated island, an average of 31 drones per virgin queen he had poor mating results, but that the next summer when he provided 59 drones per queen he had satisfactory results. It is not likely that all other mating factors were similar for both years. Brother Adam (1954) states that under special conditions he allows about 25 mature drones per virgin queen in his mating apiary, and evidently his mating results are satisfactory. Commercial queen rearers in the United States usually have several drone-rearing colonies in the apiary in order to have plenty of drones of mature age.

When a queen mates with several drones on a mating flight, as described by Triasko (1951) and Taber (1954), it is presumed that the insects find each other without much delay. Taber found the average flight time for 39 multiple-mated queens to be 13 minutes.

SUMMARY

Observations on the flight of drone honey bees (*Apis mellifera* L.) were made at Baton Rouge, La., in 1952–54. Usually the maximum number of flights was made between 2 and 3 p.m. An occasional drone was seen to fly before noon or after 4:30 p.m. A drone is likely to take more than one flight on favorable days. The flying speed of the first drone to return in each test lot ranged from 5.7 to 10 m.p.h.

The greater the distance from the hive that drones were released the fewer returned. None of those released 2½ miles from the hive returned to it. Nearly as many of the drones that were forced to fly in the forenoon were able to return as those released in the afternoon. Apparently the position of the sun is not essential as a guide. A temperature of about 60° F. probably is too low for drone flight.

Drones that had not flown averaged 220 mg. in weight, while those that returned to the hive from a flight weighed 185 mg. Drones caged as they returned from flight took 11.4 mg. of honey each during the first hour as compared with 6 mg. taken by those captured from a comb within the hive. The rate of food intake decreased with the time spent in the cage.

The removal of both or one or part of the antennae did not prevent the return of drones to the hive.

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BIOLOGY OF OLIGOTA OVIFORMIS CASEY[1] (COLEOPTERA: STAPHYLINIDAE)

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Among the many predators of the spinning mites found on citrus and avocado trees in southern California, the staphylinid beetle, *Oligota oviformis* Casey,[2] probably ranks third in importance, being outranked not only in effectiveness but also in numbers by predatory mites and *Stethorus* species. These very tiny staphylinid beetles are occasionally found in large numbers, and in such cases they contribute materially to the reduction of mite infestations. As will be shown later, the rather sporadic distribution of the insect is associated with both cultural practices and soil condition.

Both the adults and the larvae of *O. oviformis* are proficient in feeding on phytophagous mites. The larvae, as would be expected, usually consume the greater number of individuals in a shorter period of time, although the adults will take a nearly equal number of mites. Quayle (1912) showed, in the laboratory, that the feeding capacity of the larvae is considerable, each one consuming an average of 20 mites per day or, during the period of larval development, a total of from 200 to 300 mites. As for the adults, he found that their requirements numbered about 10 mites per day or, during the maximum adult life of 32 days, a total of about 320. Thus the numbers of mites eaten is about equal for larvae and adults.

Concerning pupation, Quayle (1912) stated that "repeated efforts have failed to bring it [*Oligota* larva] to the pupal stage, although our maximum records carried them to a stage just preceding pupation." The present paper describes the acts and conditions of pupation and gives detailed data on the life history and habits of *O. oviformis*.

MATERIALS AND METHODS

All observations were made in the laboratory under controlled conditions, at a temperature of 80° F. and at 50 to 60 percent relative humidity. Adults and larvae were maintained on orange fruit heavily infested with the six-spotted mite, *Eotetranychus sexmaculatus* (Riley). The infested fruits were kept in a 1-gallon battery jar, and the beetles were allowed to roam freely throughout the container. Mature larvae were tested to determine the requirements for pupation. Larvae confined in various types of cages, with or without food, died before forming pupae. Larvae confined in vials of soil immediately disappeared into the soil. These were later found to be desiccated, indicating a need for a certain minimum moisture level. A 1¼-inch Stender dish, with a well-moistened layer of plaster of Paris (treated with a fungus inhibitor), covered with a thin layer of coarse sand, proved to meet the requirements for pupation. The larvae pupated in the sand.

LIFE HISTORY AND HABITS

The egg.—When first deposited the egg of *O. oviformis* is smooth and of a pale-orange color; within a day after deposition a faint reticulate pattern is discernible. The egg is nearly oval in shape, being slightly elongated along the polar axis. Very often the egg is covered with mite exuviae, placed on it by the female immediately after deposition. The eggs may be found singly among the hosts, either in small depressions or along the midribs of leaves. Under insectary conditions the incubation period is fairly constant at 4 days.

The larva.—Almost as soon as the young larva leaves the egg it begins its search for food. At this early instar the larva is uniformly cylindrical, but as it matures it takes on the more characteristic shape described by Quayle (1912) as narrower at the anterior end and widening posteriorly. Although Quayle described the larva in some detail, he failed to mention the presence of a rather conspicuous black spot on the dorsum of the eighth abdominal segment. This black spot consists of two heavily pigmented, chitinized plates, with a transverse slit between them. The plates appear to act as a protection to an inner, bladder-like organ called the osmeterium. The larva is capable of extruding this osmeterium when disturbed. It apparently acts as a protective device, quite possibly giving off an offensive odor, as does the osmeterium of certain lepidopterous larvae.

The larva passes through three stadia, the lengths of which are variable, depending upon the availability of hosts. Under insectary conditions, with a more than adequate supply of host material, the first molt takes place within one day of hatching. The second molt takes place either the second or third day after hatching, or one to two days after the first molt. From a period of rather active feeding the larva enters a short quiescent period just prior to molting. Molting takes place in the usual manner, the old skin splitting along the mid-dorsal line of the thorax, the head and thorax being removed first, followed by the rest of the body.

The pupation site.—Usually, within 4 days after hatching or within 2 days after the last

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[2]Also referred to in the literature as *Somatium oviformis*. For the generic changes the reader is referred to: Blackwelder, R. E., 1952, The generic names of the beetle family Staphylinidae, U. S. National Museum Bul. No. 200, 271 p.

molt, the mature larvae begin searching for a place in which to pupate. If no suitable place is found within 2 days, the larvae die. Under the conditions of these studies the larvae were removed from the infested fruit and placed in the pupation cage as described above. Under natural conditions the mature larvae drop to the ground and pupate in the soil, usually not more than 1 inch below the surface. When a suitable pupation site is found the larva commences to spin a cocoon with silk secreted by glands located at the tip of the abdomen. Upon completion of the cocoon the larva enters the pre-pupal state, remaining inactive in an "S"-shaped position, with the head bent ventrad and the abdomen curved over the back. This pre-pupal stage lasts from 1 to 5 days; more commonly it lasts not more than 2 days. Therefore the total time spent in the larval stage in these experiments ranged from 8 to 13 days.

The pupa.—The exarate pupa shows the characteristic strongly deflected head and upturned abdomen of the adult. It is of a bright-orange color and, aside from a few scattered hairs on the thorax, it is naked. There are two prominent caudal cerci. As the pupae mature they turn a reddish color and just prior to emergence of the adult they are black. The exuvia of the larva remains within the pupal case but not attached to the pupa in any way. The pupal stage lasts from 9 to 13 days.

In the presence of nearly 100 percent relative humidity in the laboratory, larvae were observed to molt into the pupal stage without first spinning the cocoon. The adults which emerged from these pupae did so with a considerable amount of difficulty, and their wings and elytra were frequently deformed.

The adult.—Under normal conditions the adult is completely hardened before it emerges from the cocoon. Immediately after emerging it begins its search for food. Mating may take place as soon as the female emerges or a short time after. In the field this period may be lengthened for several days. Copulation has been observed in the laboratory to last for over an hour. A single mating is sufficient to insure fertilization of nearly all the eggs. Repeated copulation has been observed, but it is doubtful if this results in greater numbers of eggs.

The pre-oviposition period requires from 1 to 3 days, depending upon the age of the female at the time of mating. As mentioned above the eggs are laid singly in crevices or depressions and also along the midribs of leaves. Once oviposition has started the females will continue to deposit eggs for as long as 29 or 30 days. A single female is capable of producing more than 300 eggs, with as many as 16 being deposited within a 24-hour period. In one observation a single female deposited 302 eggs, of which 286 were viable.

The maximum life of the adult female under insectary conditions was 54 days. It has been observed that mated females will live from one to two weeks longer than unmated females. The length of life from the time of egg deposition until the death of the adult ranged from 61 to 84 days, the average being approximately 72 days under the stated insectary conditions.

The authors can find no reference which names the pupation site of this insect. The habit of pupating in the soil can and may have some bearing on the rather sporadic occurrence of large populations of the beetles even though the mite populations may be increasing. In southern California citrus groves the cultural practice of noncultivation very often leaves the surface of the soil hard, thus preventing the entrance of the larvae into the ground. In those groves in which frequent cultivation is practiced many of the pupae are either buried so deep in the ground that the beetles cannot emerge or left on the surface where they may become desiccated. Avocado groves generally offer better conditions for pupation and survival of the beetle. Most avocado groves are under a program of noncultivation, but because of the large number of leaves on the ground under the trees the soil remains in a rather loose condition, thus affording the larvae easy entrance and ideal sites for pupation. Consequently, *O. oviformis* is generally a more effective predator in avocado groves than in citrus groves.

Mite hosts.—The six-spotted mite, *Eotetranychus sexmaculatus* (Riley) was the mite host used as food in these studies. Other mites on which *Oligota* fed and reproduced were the citrus red mite, *Metatetranychus citri* (McG.); the two-spotted mite, *Tetranychus telarius* (Linn.); the lewis mite, *Eotetranychus lewisi* (McG.); the Pacific mite, *Tetranychus pacificus* McG.; the avocado brown mite, *Oligonychus punicae* (Hirst); the plane mite, *Oligonychus platini* (McG.); and *Tetranychus deviatarsus* (McG).

SUMMARY

Data are given on the life history and hosts of the staphylinid beetle, *Oligota oviformis* Casey, under controlled laboratory conditions at a temperature of 80° F. and at 50 to 60 percent relative humidity.

The life cycle requires from 17 to 21 days, 4 days being spent in the egg, a minimum of 4 days in the active larval stage, and from 9 to 13 days in the pupal stage. The average life span of the adult is 72 days. Pupation occurs in the soil. Females may live more than 54 days and deposit more than 300 eggs. Some mating and oviposition habits of *O. oviformis* are discussed.

REFERENCE CITED

Quayle, H. J. 1912. Red spiders and mites of citrus trees. California Agric. Expt. Sta. Bull. 234. (See pp. 509–511.)

SYNONYMY, NEW COMBINATIONS AND NOMINA NUDA IN THE GENUS EURYTOMA ILLIGER (CHALCIDOIDEA: HYMENOPTERA)[1]

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In the Catalogue of Hymenoptera North of Mexico by Muesebeck, Krombein, Townes and others, published in 1951 by the U. S. Department of Agriculture, Washington, D.C., Peck listed seventy-three species and varieties under the genus *Eurytoma* Illiger. Since that time the author has been conducting a study with the intention of revising the genus for North America. Visits have been made to the U. S. National Museum in Washington; Cornell University, Ithaca, N. Y.; and the Department of Agriculture, Division of Entomology, Ottawa, Canada to study their collections of material of the genus. Through the generous cooperation of Dr. B. D. Burks and Dr. J. F. Gates Clark at the National Museum, Dr. Henry Dietrich at Cornell, and Dr. O. Peck at Ottawa excellent facilities were made available. The following notes on synonymy, species removed from the genus, and nomina nuda are based on the material studied in the collections mentioned above and the specimens in my own collection.

The species removed from the genus *Eurytoma* were done so only after consultation with Dr. B. D. Burks who is working on a revision of the genera of the family Eurytomidae. He read the manuscript and suggested several changes which have been incorporated in the paper.

SPECIES PLACED IN SYNONYMY

Eurytoma auriceps Walsh

Eurytoma auriceps Walsh, 1870, Amer. Ent. and Bot. 2: 299.
Eurytoma auriceps seminatrix Walsh, 1870, Amer. Ent. and Bot. 2: 299. *New synonymy.*
Eurytoma seminatrix Walsh, Ashmead, 1895, Proc. Calif. Acad. Sci. [2]5: 551.
Eurytoma vagabunda Ashmead, 1881, Can. Ent. 13: 134. *New synonymy.*

There are no Walsh types of either *auriceps* or *auriceps seminatrix* for comparison purposes. The original types are believed to have been destroyed in the great Chicago fire of 1873. The original descriptions separate the two forms chiefly on the basis of differences in size and in the color of the legs. Ashmead (loc. cit.) considered them to be distinct species. Comparison of extensive series in my own collection of what is believed to be *auriceps* as well as specimens in the National Museum collection, some of which were determined by Ashmead and Gahan, has led to the conclusion that the size and color differences grade into each other and cannot be relied upon for separating the two forms.

The type of *vagabunda* Ashmead in the National Museum collection also fits into this series and cannot be clearly separated from the species I consider to be *auriceps* of Walsh. There is a curious mix-up, however, in the European literature concerning *vagabunda*. Both Silvestri 1920, (Boll. Lab. Zool. Agric. Portici. 14: 232.) and Perris 1869, (Ann. Ent. Soc. France [4] 9: 458.) refer to *vagabunda* but credit it to Giraud. Silvestri (loc. cit.) thinks it may be the same as his species *oophaga*. Actually, *vagabunda* of Giraud is a nomen nudum.

The hosts of *auriceps* and *seminatrix* are various species of gall-making *Cynipidae*.

Eurytoma pachyneuron Girault

Eurytoma pachyneuron Girault, 1916. Canad. Ent. 48: 337.
Eurytoma pater Girault, 1920. U. S. Nat. Mus. Proc. 58: 207. *New synonymy.*
Eurytoma phoebus Girault, 1920. U. S. Nat. Mus. Proc. 58: 204. *New synonymy.*
Eurytoma phoebus Girault, Gahan, 1933. U. S. Dept. Agric. Misc. Pub. 174: 33–36.

The type specimens of the first two species listed above are in the U. S. National Museum collection and can be quite easily separated. The chief difference between them is in the color of the legs. The type of *pachyneuron* has yellow fore, mid and hind coxae, and yellow fore legs. The mid legs are mostly yellow but there is a slight brownish infuscation on the tibiae. The hind legs are missing. The type of *pater* has yellow fore coxae and legs; a deeper yellow to reddish-brown mid coxae, brown infuscation on the mid femora, and deep reddish-brown hind coxae with brown infuscation on hind femora and tibiae.

The type of *pater* was described by Girault from Youngstown, Ohio and indicated as "allied with *E. auriceps*." Comparison of *pater* with the latter species must not have been actually made by Girault as the two species have very little in common beyond the characteristics typical of the genus. The type of *pachyneuron* was described from California. After examining extensive series of material bred from *Elymus* sp. in the collections of the U. S. National Museum, Cornell University, and the Department of Agriculture, Ottawa, Canada, the conclusion has been reached that Girault had specimens from the opposite ends of a cline. By following *pachyneuron* northward into Oregon and southern British Columbia and *pater* westward, the two

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species merge. Series examined from Forest Grove, Oregon and southern British Columbia contained specimens from the same collection that had been assigned to both species. There were others in the same series that fell somewhere between the two extremes.

The story for *phoebus* Girault is quite similar. Girault compared it to *vagabunda* Ashmead, but outside of belonging to the same genus, any close similarity ends. After examining the female type of *phoebus* in the U. S. National Museum collection, I do not believe that specimens, if adequate series are at hand, can be clearly separated into three species. In *phoebus* the coxae are usually all black to deep reddish-brown, although the front coxae may show yellow on the inner anterior faces. Legs are yellow with varying amounts of reddish-brown infuscation on femora and tibiae.

Color of the legs is not a dependable character, but is quite variable and in many of the extensive series examined all gradations of leg color combinations between the two extremes occurred.

Elymus species have been recorded as the host plants for the three species. In addition there are actual rearings of *phoebus* from galls of *Harmolita tritici* by T. R. Chamberlin, from *Harmolita* galls on *Elymus* by E. G. Kelley, and from Hessian fly puparia (*Phytophaga destructor* (Say)) by several collectors (see Gahan, loc. cit.). Only a few specimens of *phoebus* have been reared from the many thousands of hessian-fly puparia collected so that apparently the fly is not its typical host.

From the available evidence it would appear that the various components of *pachyneuron* are probably parasites of *Harmolita* species in various species of *Elymus*.

Eurytoma bigeloviae Ashmead

Eurytoma bigeloviae Ashmead, 1890. Colo. Biol. Assoc. Bull. 1: 25, 45.
Eurytoma chalcidiformis Girault, 1917. Desc. Hym. Chalcidoid. Variorum cum Observ. V, p. 3. *New synonymy.*

A comparison of the types of *bigeloviae* Ashmead and *chalcidiformis* Girault in the U. S. National Museum collection showed them to be remarkably alike except for a considerable difference in size. *Chalcidiformis* Girault measured approximately 5.5 mm. in length and *bigeloviae* about 3.8 mm. A third specimen turned up in the collection of Cornell University that would fall about half-way between the two. This species seems to be an inhabitant of the western half of the United States. Ashmead's specimen was described from West Cliff, Colorado. It was bred from *Trypeta bigeloviae* Ckll. Girault's came from Los Angeles, California and the Cornell University specimen from White Valley, Millard County, Utah. Neither of the latter two specimens has a recorded host.

Eurytoma orchidearum (Westwood)

Isosome orchidearum Westwood, 1869. Gardner's Chron., p. 1230.
Eurytoma phoenix Girault, 1917. Desc. Hym. Chalcidoid. Variorum cum Observ. V, p. 11. *New synonymy.*
Eurytoma phoenix Girault, 1920. Proc. U. S. National Museum. 58: 204.

Girault, in his second description of this species (1920), compares *phoenix* to *stigmi* Ashmead. These two species are quite different. When specimens of *orchidearum* and the type of *phoenix* in the U. S. National Museum were compared, however, they were found to be identical. The specimens of both *phoenix* and *orchidearum* have strongly striated faces, elongate petioles, and the median furrows of the propodeum are marked by raised lateral carinae basally. Girault recorded his specimens from San Mateo, California, from an orchid. *Orchidearum* has been bred from the bulbs, stems, leaves and buds of many species of orchids.

Eurytoma conica Provancher

Eurytoma conica Provancher, 1887. Addit. Corr. Faune Ent. Canada, Hym., p. 193.
Eurytoma phloeosini Ashmead, 1894. Trans. Amer. Ent. Soc. 21: 327. *New synonymy.*
Isosoma abnormis Ashmead, 1896. Trans. Amer. Ent. Soc. 23: 219. *New synonymy.*

I have examined the lectotype female of *E. conica* Provancher in the collection of the Department of Agriculture, Ottawa, Canada. The specimen is in excellent condition. It fits very closely the descriptions of *phloeosini* Ashmead and *abnormis* Ashmead that were made by myself from the types of the two latter species in the U. S. National Museum collection. *Abnormis* was originally described as an *Isosoma* = *Harmolita*, but was excluded from that genus in the revision of it by Phillips and Emery, 1919, Proc. U. S. National Museum 55: 433–471. The species was referred to again by C. H. Hoffman in the U. S. Dept. Agric. Misc. Pub. No. 466: 19, 1942 as *abnorme* Ashmead and listed as "reared from dead (elm) trees."

E. conica was briefly described by Provancher on page 193 of Addit. Corr. Faune Ent. Canada, Hym., but on page 438 of the same publication he listed it as a synonym of *gigantea* Walsh. If the lectotype female at Ottawa is the original specimen that Provancher described, his species is perfectly good. There seems to be no good reason to doubt this specimen. It has three labels on it; the first and second bear the name *Eurytoma conica*, with notations of type. The first label bears the number 99. The third is labeled Lectotype, *E. conica* Provancher, Comeau, April 1940. No host is indicated.

E. phloeosini Ashmead was described from a male specimen. In the U. S. National Museum collection are a number of additional specimens of both sexes. I could not find any consistent differences between the specimens of *phloeosini*

and the lectotype of *conica*. Several hosts have been cited for *phloeosini*, but among them are numerous references to rearings from elm.

Eurytoma studiosa Say

Eurytoma studiosa Say, 1836. Boston Jour. Nat. Hist. 1: 272–273.
Eurytoma succinipedis Ashmead, 1881. Trans. Amer. Ent. Soc. 9: Proc. XXXI. *New synonymy.*

Since there is no type for *studiosa* Say we have only the original description to go by. The description is so generalized that it could fit a number of species in the genus *Eurytoma*. There are several specimens in the U. S. National Museum collection determined by Ashmead as *studiosa*. Unfortunately these come from Jacksonville, Florida, some distance from Indiana, the type locality of this species. A Neotype will be picked for *studiosa* from the considerable material in my own collection from southern Indiana and Illinois. In the meantime, using material under *studiosa* in the National Museum collection for comparison, no sound morphological basis for separating *studiosa* and *succinipedis* of Ashmead, could be found.

The original description of the latter species is a very poor one also. Originally there were 4 females and 1 male, but only 2 females remain. These female specimens therefore are taken to represent the species synonymized with *studiosa*.

SPECIES TRANSFERRED TO THE GENUS EURYTOMA

Eurytoma bromi (Howard) new combination

Isosoma bromi Howard, 1896. U. S. Dept. Agric., Div. Ent., Tech. Ser. No. 2: 11.

The types consisting of two females and one male specimen, in the U. S. National Museum collection were reared from *Bromus ciliatus* at Los Angeles, California by A. Koebele. After examining the types I believe this species is a good *Eurytoma*. Gahan in Phillips, U. S. Dept. Agric., Tech. Bull. No. 518: 3 (footnote), 1936 felt the same way about *bromi* and this species was also excluded from the genus *Haromolita* by Phillips and Emery in their revision, Proc. U. S. National Museum, 55: 436, 1919.

E. bromi, in its rectangular shaped abdomen, clavate antennae, elongate first funicle joint, broad marginal vein and deep, narrow median furrow of the propodeum resembles *pachyneuron*.

Several large series of what I believe to be *bromi* were discovered in the collection at Cornell University. They were bred from the same host (*Bromus ciliatus*) as Howard's types, and collected in Ithaca, New York.

SPECIES REMOVED FROM THE GENUS EURYTOMA

Bruchophagus sculpta (Ashmead) new combination

Eurytoma sculpta Ashmead, 1887. Trans. Amer. Ent. Soc. 14: 195.

Of the four female specimens that Ashmead originally had to base his description on only one remains in the U. S. National Museum collection. The abdomen is heavily sculptured over its entire surface, the abdominal segments are all approximately equal in length, the post-marginal vein is longer than the marginal vein, and the ninth abdominal segment is very short and stubby. These characteristics are closer to the genus *Bruchophagus* than to *Eurytoma*.

Bruchophagus caraganae (Nikol.) new combination

Eurytoma caraganae Nikol'skaia, 1952. Ent. Obozrenie (Moscow) 32: 304–306.

This species has not been listed for N. America but in the collections of the Department of Agriculture, Ottawa, Canada and the U. S. National Museum are specimens, bred from *Caragana* seeds by A. Hedlin from Indian Head, Sask. Several of these specimens were determined by Dr. B. D. Burks as *Bruchophagus caraganae* (Nikol.) I was sure that they did not belong to the genus *Eurytoma*. The heavy sculpturing over most of the abdomen; the fact that the sixth abdominal segment of the abdomen is not lengthened but is about equal in length to the fifth; the almost flat, very broad propodeum with no indication of a median furrow; and the long post-marginal vein in relation to the marginal are characteristics not typical of *Eurytoma* but more so of *Bruchophagus*.

Prodecatoma maga (Girault) new combination

Eurytoma hegeli var. *maga* Girault, 1920. U. S. Nat. Mus. Proc. 59: 207.

The female type specimen of this species in the U. S. National Museum collection came from Martinez, California. It was bred from *Andricus chrysolopidicola* (Ashm.) on *Quercus dumosa* and *Q. douglassi*. A second specimen in the collection, larger than the type but resembling it closely in color, came from Descanso, Calif. It was bred from *Disholcaspis plumbella* on *Quercus dumosa*. The question as to whether this species belongs to *Prodecatoma* or *Eurytoma* depends upon the interpretation of where the anterior ocellus is located. If it is in the scrobe cavity it belongs to *Prodecatoma*, but if it is located above the scrobe cavity the species belongs in *Eurytoma*. After examining the two specimens I am convinced that the anterior ocellus is situated in the top of the scrobe cavity.

E. hegeli is a nomen nudum, as no trace of a description or further mention of such a species has been found in the literature.

Eurytomocharis muhlenbergiae (Howard) new combination

Isomorpha muhlenbergiae Howard, 1896. U. S. Dept. Agric. Div. Ent. Tech. Ser. 2: 20.
Eurytoma muhlenbergiae (Howard), Peck in Muesebeck, Krombein, Townes and Others, 1951. Hymen. Amer. North of Mexico, Agric. Monograph No. 2, U. S. Dept. Agric. p. 587.

This species was transferred to the genus *Eurytoma* as indicated above, but after examining the single specimen (female) in the U. S. National

Museum collection I do not believe it to be a true *Eurytoma*. The much posteriorly produced propodeum extends far beyond the scutellum. The median furrow of the propodeum is uniformly punctate and maintains the same width from top to bottom. These are not characteristic of *Eurytoma*. After discussion with Dr. D. B. Burks of the National Museum, who is working on a generic revision of the family Eurytomidae, the decision was made to place the specimen in the genus *Eurytomocharis*, pending the completion of the generic revision.

Ipideurtytoma polygraphi (Ashmead)
new combination

Decatomidea polygraphi Ashmead, 1894. Trans. Amer. Ent. Soc. 21: 322.
Eurytoma polygraphi (Ashmead), Peck in Muesebeck, Krombein, Townes and Others, 1951. Hymen. Amer. North of Mexico, Agric. Monograph No. 2, U. S. Dept. Agric. p. 678.

The specimens of this species in the National Museum collection were supplemented by a good series from Vancouver, B. C. in the collection of the Department of Agriculture at Ottawa, Canada. The species fits the description of the genus *Ipideurytoma*, described by Bouček and Novicky, 1954, Entom. Tidskrift, Agr. 75(2–4): 266–269, in which *spessivtsevi* from the vicinity of Stockholm, Sweden, is described as a new species as well as the genotype. *I. spessivtsevi* was bred from second generation larvae of *Xyloterus lineatus*. *I. polygraphi* is listed, questionably, as bred from *Polygraphus rufipennis* (Kby.) in Cat. Hymen. (loc. cit.). The specimens from Vancouver, B. C. referred to above, are labeled, "Host, *Trypodendron*." *I. polygraphi* has been shunted about, having originally been placed in the genus *Decatomidea* established by Ashmead, 1888, Ent. Amer. 4: 42, 43. *Decatomidea* is listed as being synonymous with *Eudecatoma* Ashmead in Cat. Hymen. p. 579, 1951, and *polygraphi* was referred to the genus *Eurytoma* (loc. cit., p. 578). *I. polygraphi* with its small, plump abdomen, short and barely exposed dorsal valves, and its broad depressed pronotum that is produced forward dorsally as a broad rounded lobe, thus becomes the second species in the new genus *Ipideurytoma* of Boucek and Novičky.

NOMINA NUDA

Eurytoma hegeli Girault. The only mention of this species is in a paper by Girault, (Proc. U. S. Natl. Mus. 58: 207, 1920) in which he describes the variety *maga* of *E. hegeli*. Since no description or other reference to this species seems to exist it would appear to be a *nomen nudum*.

Eurytoma albitarsis is credited to Ashmead by two different authors, but no description or specimens can be found. Chittenden, 1902, U. S. Dept. Agric., Div. Ent. Bull. 38 cites *E. albitarsis* Ashmead as a parasite of *Microrhopala xerene* (Newn.) in *Solidago* in the vicinity of the District of Columbia. McCauley, 1938, Bull. Brooklyn Ent. Soc. 33(4): 145–168 cites the same parasite and host species, indicating Chittenden's publication as his source. I have been unable to find any way in which Chittenden could have obtained this name unless it had been applied by Ashmead to specimens submitted to him for identification. A description was apparently never published.

Eurytoma aulacis is mentioned by Brodie, 1892, Can. Ent. 24: 12–14, and credited to Ashmead. Brodie records *Aulax nabali* Brodie as its host and its locality as Toronto, Canada. No previous or subsequent reference to this species has been discovered, and so far as could be determined there are no types or description. *E. aulacis* appears to be a *nomen nudum*.

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Scientific names of insects and other animals should be checked for accuracy and completeness. The author of every species mentioned should be given, in parentheses if necessary, at least once in the text of every manuscript. Scientific names of plants should be given in conformity with accepted botanical usage. In the presentation of taxonomic material, the author should consult the pages of the ANNALS for accepted usage and should conform with the style in so far as it is adaptable to his particular needs of presentation.

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ERRATUM

In the ANNALS, vol. 49, no. 3, p. 236, col. 1, line 6, the botanical name *Nepeta hederacea* is incorrectly given for the English Ivy. The proper name is *Hedera helix*. According to the authors (DeBach and Fisher) this prostrate ivy is locally called ground ivy, an accepted common name for *Nepeta hederacea*.

The following Tables are reprinted here because the asterisks referring to the footnotes were omitted from them in the article by Stanley D. Beck entitled, "Nutrition of the European Corn Borer, *Pyrausta nubilalis* (Hbn.). IV Feeding Reactions of First Instar Larvae," which appear in Volume 49(4): 399–405. 1956.

TABLE III

THE EFFECTS OF PLANT SUBSTANCES OF DIFFERENT CORN LEAF FACTOR ACTIVITY ON THE ESTABLISHMENT OF FIRST INSTAR EUROPEAN CORN BORER LARVAE

Experiment	Dietary adjuvant	Larval establishment (%)	Corn leaf factor content of adjuvant[a]
1 (Four-choice)	None(control)	20.0	——
	Leaf sap concentrate	3.4**	high
	Corn leaves	34.6*	high
	Corn tassels	42.0**	high
2 (Two-choice)	None (control)	20.5	——
	Aqueous extract of corn leaves	79.5**	high
3 (Two-choice)	None (control)	36.7	——
	Brewer's yeast powder	63.3*	low
4 (Two-choice)	None (control)	34.0	——
	Petroleum ether extract of corn leaves	66.0**	none

[a]As reported by Beck (1953)
* Significantly different from control response.
** Highly significantly different from control response.

TABLE II

THE EFFECT OF DIFFERENT AMOUNTS OF THE COMPONENTS OF THE BASAL DIET ON THE ESTABLISHMENT RESPONSE OF FIRST INSTAR LARVAE OF THE EUROPEAN CORN BORER

Nutrient varied	Concentration A	Larval establishment at indicated concentration			
			Concentrations		
		control[a]	A	2xA	4xA
	(mg/diet)	(%)	(%)	(%)	(%)
Glucose	100	4.7	20.0*	26.1**	49.2**
Casein	1000	15.6	21.6	30.5*	32.3*
Corn oil	20	20.4	24.0	24.9	30.7
Cholesterol	30	20.5	25.6	25.2	28.7
Wesson's salts	75	23.8	16.9	33.1	26.2
Choline chloride	10	26.2	29.0	22.1	22.7
B-Vitamin mixture	10	26.4	24.3	27.1	22.2
Brewer's yeast powder	300	12.8	23.8*	28.4**	35.0**
Leaf factor concentrate	200	68.5	16.8**	10.5**	4.2**

[a]Test nutrient omitted.
*Significantly different from control response.
**Highly significantly different from control response.

THOMAS SAY FOUNDATION, VOLUME II

A supply of Volume II, *Plecoptera or Stoneflies of North America*, by J. G. Needham and P. W. Claassen has recently been located. This makes all five volumes available that have been published by the Foundation. Copies of Volume II sell for $5.00 postpaid in North American and U. S. Possessions, and for $5.50 elsewhere.

BOOK NOTICES

LES ANOPHELES DE L'AFRIQUE DU NORD ET DU BASSIN MEDITERRANEEN, by G. SENEVET and L. ANDARELLI. Paper. 280 pp., 6 pls., 67 figs. P. Lechevalier, Paris. 1955. 3,000 francs.

The authors of this work, chiefs of the laboratory of the Pasteur Institute of Algeria and the Anti-malaria service of Algerie respectively, have treated the *Anopheles* species of North Africa and the Mediterranean region in careful detail. The first part, of 28 pages, deals with the general anatomy of all stages of *Anopheles*. The second part, of 169 pages, deals with the 17 species. Each species is described and illustrated, with notes on variations, biology, and relation to malaria. The races of *Anopheles maculipennis* are discussed at considerable length, and to the great confusion of a cataloguer. We find on page 71 a list of the races or varieties of *maculipennis* with no indication as to which are races and which are varieties. To take one of the names, *labranchiae*, we find on page 71 it is treated as a race (or subspecies), on p. 82 as a full species, *A. labranchiae*, and on page 278 as *A. maculipennis* var. *labranchiae*. The taxon *atroparvus* is treated as a race of *maculipennis* on p. 71, as a subrace of *labranchiae* on page 82, and as a variety of *maculipennis* on p. 278. We can derive from this work the combination *Anopheles maculipennis labranchiae labranchiae sicaulti* if we combine the concept expressed on page 71 with that on page 82. This reflects a confusion in the minds of the authors as to what a species, a race, and a variety are.

The third section gives detailed distribution records for all of the countries covered, and three sets of keys; one to all of the species included in the work, one to the species of France, and one to those of the Mediterranean region. This is a very useful work to anyone engaged in studies involving the Anopheles mosquitoes of southern Europe or North Africa.

ALAN STONE.

BIOTA. Instituto Salesiano "Pablo Alberta", Magdalena del Mar, L. Prado 520, Peru. Editor, J. Soukup, S.D.B.

The (apparently) first volume of this new publication has been received. It consists of four numbers, the last two forming a double number, with a total of 94 pages. The dates are October, 1954, through August, 1955. There is no statement of policy or objectives, but the ten articles of this volume deal with the animal and plant life of Peru, chiefly from a systematic standpoint. Of these articles, six are botanical; two deal with echinoderms and one with Odonata (a catalogue of the Peruvian species). The tenth is biographical, commemorating the completion of twenty-five years of scientific collecting in Peru by Felix Woytkowski. This will be of particular interest to North American biologists who have used Woytkowski's material, particularly in view of the fact that the collecting localities are listed and grouped according to ten collecting centers. A "Bibliography for Peru" and a "Directory of specialists" are being run serially; the object of the latter is stated as being "to put into communication the greatest possible number of specialists in order to facilitate the classification of plants and animals". A request for names is made.

MAURICE T. JAMES.

AFRICAN INSECT LIFE, by S. H. SKAIFE. Pp. viii+387, 190 figs., 70 plates (black and white), colored plates. Longmans Green and Co., New York. Price $11.00.

In its approach, this book is intermediate between the more technical introductions to entomology on a college level and the various popular treatments of the subject. It fills, consequently, an interesting niche which will make it valuable to both the entomologist and the more serious minded non-professional. The approach is scientific, yet at the same time it avoids technicalities, both in subject matter and in language. For example, for descriptive purposes wing venational characters and genitalic structure are not used, though such characters as a "feathered" arista and "three-jointed tarsus" are. No keys are given, and classifications (including suborders and the more important families, with a brief characterization of each) are appended to the individual chapters.

The book begins, without an introduction (except for the brief preface), with the "primitive insects" (Chapter I), and most of the subsequent twenty-one chapters are devoted each to a single order of insects (the last three, however, taking up the wasps, bees, and ants separately). There are, consequently, no separate sections devoted to insect anatomy, life histories, and such; the basic anatomical material may be found in chapter II, under "cockroaches", and life histories of the various members and orders are given in their respective places in the text. There is no glossary; the addition of one would have helped considerably to enable the work to stand by itself, particularly in view of the fact that such terms as "larva", "pupa", and "metamorphosis", though used throughout the text, are either omitted from the index or are listed there in references which do not include definitions. The author probably considered that any reader capable of using the work could get such information from a dictionary. A brief list of references would have been useful.

The book is replete with interesting material, much of it of common knowledge to the professional entomologist, but much of it obviously gleaned from less well-known sources. Lack of documentation makes it difficult to determine how much of the material is original, but obviously quite a little material has come from the experiences of the author or of his colleagues. The illustrations are good and for the most part adequately reproduced. The book can not replace such a text as Imms' "Textbook of Entomology", though it can well be used by students and teachers as a collateral work. It is intended particularly "for the amateur student of natural history, for the farmer, gardener, and householder who wish to know something about the numerous insect friends and foes they see about them" (Preface). For such a reader group "African Insect Life" occupies a unique position.

MAURICE T. JAMES.

THE ARMORED SCALE INSECTS OF CALIFORNIA, by HOWARD L. MCKENZIE, Bulletin of the California Insect Survey, vol. 5, 1956, pp. 209, illus. (some col), University of California Press, Berkeley and Los Angeles, Calif., paper $4.50, cloth $6.00.

This volume covers the 132 armored scales that are now known to occur in the State of California, presenting keys for their recognition, illustrations showing details of structure, and, for most of the species, detailed information on distribution within the state and on host associations, this information presented in part through a table showing distribution by counties and through a separate plant host list organized by genus under each plant family.

The classification and keys presented adhere closely to the system proposed by G. F. Ferris in his 1942 volume of the Atlas of North American Scale Insects and are therefore well known to workers on the Coccoidea. Many of the included drawings of structural details of species have been derived from the Ferris Atlas volumes, but they have been supplemented as necessary by illustrations by the present author.

Two things stand out strikingly in the volume: The first is the very successful use of reproductions of colored

photographs to present the habitus appearance of 27 of the more important California armored scales; the second is the assignment of a common name to each of the species treated in the volume. A few lapses have been noted, perhaps the most important of which is the use of "ventral" for "dorsal" at the end of the first line of the second couplet in the key to the subfamilies, p. 21.

In view of the economic importance of these insects in California—introduced or apparently introduced species, including many serious pests, considerably outnumber indigenous ones—it seems certain that this volume will prove very useful to working entomologists not only within that state but throughout the western part of the country.

HAROLD MORRISON.

A REVISION OF THE GENUS MEGARTHROGLOSSUS JORDAN AND ROTHSCHILD, 1915, (SIPHONAPTERA: HYSTRICHOPSYLLIDAE), by EUSTORGIO MENDEZ. University of California Publications in Entomology. Vol. 11, No. 3, pp. 159–192, 14 figures in text. Published July 26, 1956 by University of California Press, Berkeley 4, California. Price: $.85.

This paper is a thorough and carefully done study of the anomiopsylline genus *Megarthroglossus* Jordan and Rothschild, which occurs only in western United States and Canada. *Megarthroglossus* species are highly modified fleas adapted for existence in the nests of rodents. Mr. Mendez is to be congratulated on the quality of his illustrations, and the only improvements might have been addition of entire drawings of both male and female modified segments.

Flea workers will welcome this addition to the steadily growing list of generic revisions within the order Siphonaptera.

PHYLLIS T. JOHNSON.

HISTOLOGY OF THE OVARY OF THE ADULT MEALWORM *TENEBRIO MOLITOR* L. (COLEOPTERA, TENEBRIONIDAE), by LOREN L. SCHLOTTMAN and PHILIP F. BONHAG, University of California Publications in Entomology, Vol. 11, No. 6, pp. 351–394, pls. 42–50, Sept. 14, 1956. Price $1.00.

The morphology and histochemistry of the ovarioles and the development of follicles are discussed. The telotrophic ovariole of *Tenebrio* is compared with the similar ovariole of the Heteropteran *Oncopeltus fasciatus*. Localizations of ribonucleic acid (RNA) and desoxyribonucleic acid (DNA) are given, along with evidence of the contribution of these acids by the apical trophocytes to the enlarging oocytes. It is interesting to note that the authors believe the follicular epithelium develops from the interstitial cells of the germarium via the prefollicular tissue. Protozoan parasites in the ovarioles were quite common. Good photomicrographs show cellular structure, protozoan parasites, and evidence of the acids mentioned.

T. J. SPILMAN.

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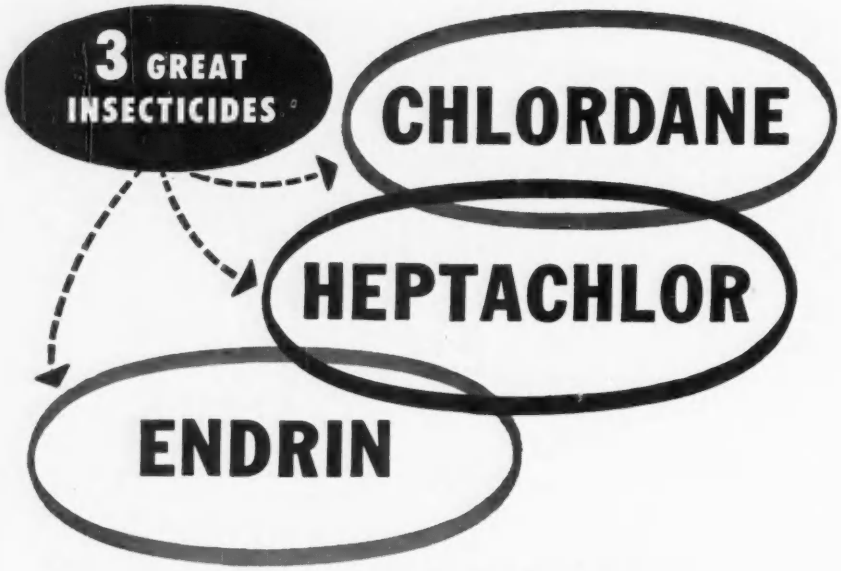

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